

PUBLISHED BY
THE EUROPEAN AND MEDITERRANEAN CEREAL RUSTS FOUNDATION

EDITED BY R.A. BAYLES

VOLUME 20 PARTS 1 & 2, 1992

CEREAL RUSTS
AND
POWDERY MILDEWS
BULLETIN

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EFFECT OF EARLY POWDERY MILDEW INFECTION ON BARLEY UNDER SIMULATED
DROUGHT STRESS

F. ZINE ELABIDINE, M. REINHOLD AND A.L. SCHAREN

U.S. Department of Agriculture Research Service and the Department of
Plant Pathology, Montana State University, Bozeman 59717.

ABSTRACT

The effect of powdery mildew on barley shoot and root development was studied. The disease was found to cause a reduction in size and number of shoots and of both adventitious and seminal roots. However, the effect on roots was more striking. When inoculated and healthy plants were subsequently subjected to different soil-water levels, the effect of powdery mildew was more damaging. Inoculated plants wilted sooner than healthy plants depending on the amount of available water. Shoots and roots of inoculated plants exhibited a larger decrease in dry weight, number and size when compared to healthy plants. It is concluded that barley seedlings infected with powdery mildew are more vulnerable to damage caused by late season drought.

Barley (*Hordeum vulgare* L.) powdery mildew is a severe disease that occurs wherever the crop is grown. The causal agent, *Erysiphe graminis* DC. f. sp. *hordei* Em. Marchal, lives superficially by sending haustoria into epidermal cells of aerial parts of the plant.

The importance of the disease for the temperate humid climate regions of the world is well documented (last, 1962, Brooks, 1972, Jenkyn, 1974, Griffith et al, 1975, Jenkyn and Bainbridge, 1978). In many semi arid climates of the world however, powdery mildew symptoms are only observed early in the growing season. In several Mediterranean countries for example, barley powdery mildew only recently has been recognized as a damaging disease. Few studies have been conducted in this particular environment (Schluter, 1976, Ghobane et al 1981, Skorda, 1981) indicating a disastrous effect of early infection on the crop. Losses of 10 to 30% have been reported for North Africa. The disease seems to be of equal importance in Mediterranean Europe. Near the Mediterranean Sea, barley is produced under many climatic and edaphic conditions. The crop is often grown on marginal lands, and under poor crop management subjecting the plants to many additional stress factors (Srivastava, 1977).

Water stress affects barley growth and development. The critical stages requiring adequate soil-water conditions are tillering, anthesis and heading stages. At these growth stages water stress reduces yielding ability of barley plants (Weltzien and Srivastava, 1981).

To study the combined effect of powdery mildew and water stress, a 2x4 factorial, completely randomized design with four replications was used. The two factors studied were: 1) treatment (inoculated, healthy controls), and 2) water levels (0 = no water, 1/3 of normal water = 1 L daily, 2/3 of normal water = 2 L daily, and 1 = 3 L daily). Each replication contained 32 plants (16 inoculated, 16 healthy). Sixty days after plant emergence at the beginning of stem elongation,

A completely randomized experimental design, employing one seedling per pot as the experimental unit, with two treatments (inoculated, healthy controls) was used to study the effect of powdery mildew on barley shoots and roots. Three experiments were conducted consecutively, using 14, 12, 13 replications per treatment, respectively. Thirty days after planting when the plants had developed several tillers, the roots were washed thoroughly under running water. Shoots and roots were dried separately for 12 hours at 70°C, then weighed. A one-way analysis of variance was performed to compare dry weight of shoots and roots.

Plants were inoculated at the 2 - 3 leaf stage about ten days after emergence by shaking or brushing conidia from diseased plants onto healthy leaves. Leaves of diseased plants were periodically disturbed throughout the duration of the experiment. This resulted in a heavy disease development on the entire plant. The disease severity was estimated with 70 percent of leaf area covered. The isolate used was Bz1, collected near Bozeman, Montana.

Plants were grown in a mixture of 3 parts sand, 1 part soil (v/v) in plastic pots (9x9 cm). Two seeds were planted per pot and the seedlings were later thinned to one per pot. Plants were kept in a growth chamber under finely woven muslin cages (50 x 61 x 76 cm) with a 12 hr photoperiod (2.2-3.3 x 10 ergs cm⁻² s⁻¹ (1 erg = 0.1 J)) at 15:24 + 1 C (dark:light). Average relative humidity was 80%. Cages were fitted in metal pans (1.25 x .55 x .025 m each) to facilitate watering (3 L/pan/day). Once a week each plant received 45 ml of the following nutrient solution: Ca(NO₃)₂·4 H₂O, 0.850 g; NaNO₃, 0.185 g; KH₂PO₄, 0.164 g; KNO₃, 0.144 g; MgSO₄, 0.236 g; H₂O, 1000 ml.

The barley cultivar Manchuria (CI 2330), susceptible to all known races of *E. graminis hordei*, was used throughout this study to produce the inoculum.

MATERIALS AND METHODS

Only a few authors have investigated the effect of powdery mildew and drought on plant growth. Ayres and Zadoks (1979) studied the effect of the disease on barley plants grown and maintained at three different soil-water levels. They found that lack of soil-water and powdery mildew had additive, deleterious effects on barley growth and yielding ability. In order to shed further light on interactions of powdery mildew and drought, the objectives of this investigation were: 1) to study the effect of powdery mildew on barley shoots and roots, and 2) to study the effect of both powdery mildew and drought on barley plants.

In this investigation, early infection by powdery mildew and water stress had an additive and harmful effect on barley plant growth. Because powdery mildew infection induces a reduction of root size, weight and mass, the impact on barley yield may be devastating (Ayres and Zadoks, 1979, Brooks, 1972, Griffiths *et al*, 1975, Last, 1962). Deep roots and abundant root hairs are critical when soil-water is lacking or unevenly distributed (Mackey, 1980). Plants resisting

In this study, powdery mildew was found to cause a significant reduction of both shoot and root growth of infected seedlings. Root size, mass, and dry weight were decreased. This reduction was first observed when soil was washed from roots. Similar results were reported by Last (1962), Brooks (1972), and Griffiths *et al* (1975). Dry weight of shoots was also reduced. A reduction in the dry weight ratio of root/total plant was observed. These effects on roots may be attributed to early and severe infection by powdery mildew. The devastating effect of early powdery mildew infection on subsequent plant development has been reported by Lim and Gaunt (1986a,b). Slowing of shoot growth was generally proportional to reduced size of roots of the plants (Last, 1962).

DISCUSSION

When both powdery mildew (disease) and reduction of water (simulated drought) were applied simultaneously to barley plants, the dwarfing effect on shoots and roots was increased well beyond the effect of either stress alone (Table 1). The maximum reduction in weight of roots was 90% and reduction of shoots was 91%. The ratio of root to total plant was affected mainly by the disease (Table 2). Water levels affected the ratio little, if at all.

Reduction in the amount of water applied, thereby putting the plants under moisture stress, also caused diminution in the size of shoots and roots (Table 1). In the uninoculated treatments, roots under maximum moisture deprivation were reduced 66% and shoots 57% when compared with those having a normal water supply.

Heavy infection with *E. graminis* f.sp. *hordei*, causal agent of the powdery mildew disease, resulted in substantial and significant reductions in weight of both shoots and roots of barley plants (Table 1). On a percentage basis, shoots were reduced about 66% and roots about 76% when compared with non-inoculated controls. In inoculated treatments, shoots were stunted, while roots were thin and sparse compared with the checks.

RESULTS

The roots were washed 15 days after water treatments were applied. The roots were washed 15 days after water treatments were begun in the first and second experiments. In the third experiment, roots were washed 21 days after water treatments were applied. Plants receiving no water were visibly wilting at this time. Data from the three experiments were combined and analysed by a multi-factor analysis of variance.

powdery mildew infection and developing healthy normal root systems should be able to make better use of available water. In our experiments, powdery mildew infection reduced the size of roots about equally regardless of the amount of water supplied to the barley plants. Soil-water stress may determine the magnitude of the losses induced depending on the drought severity and timing. The most critical stages of barley are tillering and heading (Weltzien and Srivastava, 1981). The effect of the fungus and drought on grain yield could not be determined, since the experiments were completed before heading stage. Ayres and Zadoks (1979) reported that powdery mildew caused a reduction of grain weight, and drought reduced the number of grains per ear.

Our results should be of particular significance to the many regions of the world where barley is grown under less than optimal conditions. Especially in these areas, the use of varieties resistant to powdery mildew and tolerant of drought may increase barley production. Such varieties should escape the devastating effect of early infection by the fungus, and should have a better chance of tolerating the later occurring water stress.

ACKNOWLEDGEMENTS

The authors gratefully acknowledge the technical assistance of MaryEllen Dietz-Holmes, Robin Wooding and Frank Caplette. Financial support of USAID-ICARDA Contract AID/DSAN-C-0024 is also acknowledged.

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Table 1. Dry weight of barley shoots and roots, inoculated with *E. graminis* hordei and with four levels of watering.

Water level ^a	Dry weight, g ^b			
	Shoots		Roots	
	Healthy	Inoc.	Healthy	Inoc.
1	0.610 (100)** ^c	0.205 (34)**	3.38 (100)**	0.81 (24)**
2/3	0.430 (71)**	0.093 (15)**	2.49 (73)**	0.72 (21)**
1/3	0.305 (50)**	0.074 (12)**	1.51 (45)**	0.61 (18)**
0	0.258 (42)**	0.053 (9)**	1.16 (34)**	0.35 (10)**

a Normal watering (level 1) was 3 litres per experiment per day
 b Shoot and root weights are means of three experiments
 c Means were significantly different at P<.01.

Table 2. Ratio of root/total plant in barley inoculated with *Erysiphe graminis* hordei and with four levels of watering.

Water level ^a	Healthy	Inoculated
1	.85	.20
2/3	.85	.11
1/3	.83	.11
0	.82	.13

a Normal watering (level 1) was 3 litres per experiment per day
 b Shoot and root weights are means of three experiments

PREVALENCE AND DISTRIBUTION OF PHYSIOLOGIC RACES OF STEM RUST
(PUCCINIA GRAMINIS TRITICI) IN PENINSULAR INDIA DURING 1989 TO 1991

R J PATIL, R T SAPKAL, J V PATIL AND B K PATIL
Regional Wheat Rust Research Station, Mahableshwar-412 806, India

SUMMARY

The analysis of stem rust samples collected from Tamil Nadu, Karnataka, Maharashtra and Madhya Pradesh revealed that the complexes R 40, 40A, 15C, 21A1 and 21A2 of stem rust (*Puccinia graminis tritici*) were prevalent during the years 1989 to 1991.

INTRODUCTION

Stem rust caused by *Puccinia graminis tritici* is the most damaging of the wheat rusts. It is an important disease in Peninsular India. Survey and analysis of samples is, therefore, essential to forecast the preventive measures for farmers, to use resistant varieties and also to scientists to deploying the genes for developing rust resistant varieties against the identified races.

MATERIALS AND METHODS

Stem rust infected samples were received from different States, viz. Tamil Nadu, Karnataka, Maharashtra and Madhya Pradesh. These were established on a susceptible cultivar "Pusa-4" in a glasshouse. They were analysed on a set of International Differentials (Stakman et al., 1922) and on 'OAB' set as per Bahadur et al., 1985.

RESULTS AND DISCUSSION

The prevalence and frequency pattern of different races of stem rust in the States are presented in Table 1. This study revealed that race complex 40A and 21A2 of stem rust occurred at the highest frequencies.

Races 40 and 40A were detected from Tamil Nadu during 1989 to 1991. Races 40, 40A and 21A1 were prevalent in Karnataka. In Maharashtra, Races 15C and 21A2 were prevalent while race 40A was prevalent in Madhya Pradesh.

During 1981-82, More et al (1985) reported races/biotypes 15C, 21, 21A1, 34A, 40A1, 117, 117A, 117A1 and 122 from Peninsular India.

During 1984-85, Mutkakar et al (1987) also reported the rice flora of Peninsular India, races/biotypes 15C, 40, 40A, 42B2, 117, 117A and 122 were predominant in nature.

The present race analysis indicated that race complex 15C, 40, 40A, 21A1 and 21A2f of stem rust occurred at highest frequencies during 1989 to 1991 and suggest the need for developing rust resistant varieties against above races.

ACKNOWLEDGEMENT

Authors are thankful to the Wheat Rust Mycologist, Regional Wheat Rust Research Station, Mahableshwar for providing facilities for above research.

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Table 1. Prevalence and per cent frequencies of races and biotypes of stem rust (Puccinia graminis tritici) in Peninsular India during 1989 to 1991.

Stage	1989-90		1990-91	
	No. of samples analysed	Race detected	No. of samples analysed	Race detected
1. Tamil Nadu:				
(a) Off season	51	40A(52.94%) 40 (31.37%) 40 & 40A(15.68%)	53	40A(62.26%) 40 (37.73%)
(b) Rabi season	-	-	-	-
2. Karnataka				
	6	40 (100%) 40A (72.30%) 21A1(9.23%)	65	
3. Maharashtra				
	5	15C(100%)	7	21A2(71.42%) 40A(28.58%)
4. Madhya Pradesh				
	3	40A(100%)	1	Inoculum not established

Figures in parentheses indicate percentage of frequency of prevalent races/biotypes.

The seedlings of the wheat varieties were grown in four inch diameter earthen pots in the glasshouse. Seven day old seedlings were transferred to the testing room of the glasshouse and inoculated with individual physiologic races/biotypes of stem rust. Inoculated pots were exposed to higher humidity in moist chambers for 24 hours and then transferred to glass house benches for development of rust. Observations on the type of pustules were recorded 16 days after inoculation as per the key prepared by Stakman and Levine (1922). The average maximum and minimum temperatures during the crop growth were between 8.7°C and 26.6°C with relative humidity 57 per cent.

Twenty five varieties of wheat were sown in the field (1988) at Regional Wheat Rust Research Station, Mahabaleshwar, representing each entry by one row of 1 metre length. The inoculum of a mixture of stem rust races was sprayed on the plants to create artificial epiphytotic. Observations on per cent severity and resistant/susceptible reactions were recorded as per the modified Cobb's scale (Peterson et al., 1948).

MATERIALS AND METHODS

Testing of a large number of wheat varieties against different physiologic races/biotypes is a pre-requisite for breeding rust resistant lines. The success of any breeding programme depends primarily upon the availability of a large number of resistant donor parents. A variety susceptible in the seedling stage may or may not remain susceptible in the advanced stage of growth (Goluden et al., 1928) and hence, resistance in both the stages need to be ascertained. Stem rust of wheat causes enormous crop losses in quality and quantity of produce when it appears in an epidemic form. Screening of genetic sources therefore is an important aspect in developing new varieties possessing good agronomic characters combined with good disease resistance.

INTRODUCTION

The study showed that entries DL 230-5, HI 977, HS 191, HD 2402, K 8228 and VW 147 were resistant against stem rust (*Puccinia graminis* f.sp. *tritici*) of wheat as they conferred resistance in seedling as well as in adult plant stages.

SUMMARY

Regional Wheat Rust Research Station, Mahabaleshwar 412 806 (India)

R.T. SARKAL, R.J. PATIL and J.V. PATIL.

SOURCES OF RESISTANCE TO STEM RUST OF WHEAT (*Puccinia graminis* f.sp. *tritici*)

Cereal Rusts and
Powdery Mildews Bulletin

Vol. 20, Parts 1, 2 1992

RESULTS AND DISCUSSION

The data presented in Table 1 revealed that out of twenty five varieties; DL 230-5, HI 977, HS 191, HD 2402, K 8228 and VW 147 showed resistance, to all the races and biotypes in seedling and against the mixture of races in adult plant stages.

The varieties CPAN 1796, HB 208, HB 629, HD 2379, K 8152, BW 120, BW 145, Raj 2848 and HW 953, although susceptible to a few races/biotypes at seedling stage, were found resistant at adult plant stage, indicating that entries having resistance at adult plant stages do not necessarily possess seedling resistance and vice versa. Therefore, it may be concluded that seedling resistance and adult plant resistance are quite distinct. This observation is in confirmation to that reported by Goluden *et al.* (1928) and Mutkekar *et al.* (1985).

The entries HD 2403, HS 176, K 8622, BW 117, BW 124, BW 138, UP 1109, UP 1110, VL 421 and VL 639 were susceptible at seedling as well as adult plant stages. Parentages of the entries tested against stem rust of wheat are given in Table 2.

ACKNOWLEDGEMENT

Thanks are due to Principal Investigator (Wheat Pathology), I.A.R.I., New Delhi for supplying the seed of entries studied in the present investigation.

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Table 1: Seedling and adult plant reactions of the wheat varieties against stem rust of wheat.

Variety	Race/biotype															Field Reaction		
	11	14	15	15C	21	21A	21A1	24	24A	34	40	40A	42	117A1	122		184	295
1. CPAN 1796	2	0	2	1	0;	4	0;	0	0;	1	2	0	0	0;	3	0	0	10 MR
2. DL 230-5	0	2GI	2	2	0	2GI	1-2	2	0	1-2	0	0	0;	2GI	2	0	2	40 MR
3. HB 208	2	0;-1	2	2	2GI	2GI	0	0	0;-2	0	2	0	4	2	0	0-2	2	20 MR
4. HB 629	0	1-2	2	3	2+	0	4	0	0	0	1	0	4	0-2	0	2	0	30 MR
5. HD 2379	0	2+	2GI	2	1-2	2GI	2GI	2	2	2	0	1	3	2	0	1	2+	20 MR
6. HD 2403	1	2:	2GI	2	1	0	1-2	2	3	1	1	0	2	0	2	0	3	Trace's
7. HI 977	1	2	1-2	1-2	2GI	2GI	2GI	2	1	1	1	0	2+	2	2	2-	0	30 MR
8. HS 176	3	3	3-4	3	2	4	1-2	3	0	3	0	0	3	0;	0	0;	4	20 MR, 20's
9. HS 191	1	0	1-2	0;-1	0;	0	0;-1	0	0;	1-2	2+	0	0	0	1	1	2	40 MR
10. HD 2402	2	2	1	2	-	2+	0	2	0	0	0;	0	0	2	0;	1	2	30 MR
11. K 8152	1	3	1	2GI	2GI	2GI	2GI	3	2+	-	0;-1	0	0	2	0;	2	4	30 MR
12. K 8228	0	0;	2+	0;	-	2+	0;-1	0;	0	-	0;	1	0	1	2	1	2	Free
13. K 8622	2	1-2	0	2	-	3	0	0	1	0;	0	0	2	2GI	2	2-	0	Trace's
14. PBW 117	2	3-4	2	3	3	4	1-2	4	3	0	0;-1	1	2	4	2	1	3	80's
15. PBW 120	1	2GI	1-2	2	0;-1	3	2	2	2	2	1	1	2	2GI	3	1	2	60 MR
16. PBW 124	2	0	1	2GI	0	3-4	2GI	0	0	2	0	0	0;	2	4	0	4	40's
17. PBW 138	2	0	0	1-2	0;-1	2GI	2	2	2	0;	0	0	0	2	0	3-4	0	40's
18. PBW 145	2	2GI	3-4	2GI	3	2GI	2	1	2GI	2	2	0	2+	2	0	0;-1	2GI	30 MR
19. HW 953	2	1	2+	2	2-4	2GI	0;	1	2GI	0	1	2	4	2GI	2	0	2+	50 MR
20. Raj 2848	2	2	2	2	2	0;-2	2GI	2	2GI	2	2	0	3	2GI	2	0;	2GI	Free
21. UP 1109	0	0	3	2	0	4	0	0;	0;	-	2+	0	1	1	0	0	3-4	5's
22. UP 1110	1-2	2++	2	2	2	4	1	1	2	2	1-2	0	1	2GI	4	0	0	20 MR, 10's
23. VL 421	2	0;	3	2	0	4	1-2	0	0	2	2	0	0;-1	1	4	0	4	10's
24. VL 639	0	2	2GI	0	2	-	3	2	0	2+	2	0	0	3	0;	0;-1	2GI	20 MR, 10's
25. VW 147	0	0;-1	0;-1	2	0;-1	2GI	0;-1	2	0;-1	2++	1-2	0	0	1-2	2	0	2	Free

T-Trace, MR-Moderately resistant, S-Susceptible,
R-Resistant, MS-Moderately Susceptible, GI-Green Island.
0-Immune, 0,-Very Resistant, 1-Resistant,
2-Moderately Resistant, 3-Moderately Susceptible,
4-Susceptible

Table 2 Percentage of the entries tested against stem rust of wheat

1.	CPAN 1796	Npo-Tob"s" x 8156/Kal-Bb CM 7806-15M-2y-2M-0M
2.	DL-230-5	K 7537 x HD 2160 Mut
3.	HB 208	E4717 x HB (M) 65-50
4.	HB 629	Rj 62-Gallo/Nr x Flr-Cno
5.	HD 2379	HD 2275 x HD 2289
6.	HD 2402	(HD 2267 x HD 2236)
7.	HD 2403	HD 2267 x HD 2254
8.	HI 977	(Gallo-Aust-61-157) x Cno.No.66-Y50E-Kal.
9.	HS 176	7c Nad 63 x Cno 64
10.	HS 191	HB 234 x HD 2119
11.	HM 953	CMM 268 (Gallo-Aust-11-61 157 Cno-Nor-56) x RR68 (WM15/Jis Cno x No. 68)
12.	K 8152	Tzpp ² -ANE ³
13.	K 8228	Meng 8165 x Jar's'/Cy(cad)
14.	K 8622	C.C. 505
15.	PBW 117	G 11's' - BYE ² - 7C*(ZBxM)X DML 5001
16.	PBW 120	WG 377-HD 2160
17.	PBW 124	Pichihula-WL 395
18.	PBW 138	Ravi 43-HD 2177
19.	PBW 145	HD 2160-WG 1025
20.	Raj 2848	HD 2160xHD 1981
21.	UF 1109	UF 262/UF 368
22.	UF 1110	(Bb-Tob)(Cno-No.66/C 273xNP 875 E 853) ² B)/UF 270
23.	VL 421	Son. 64 x Y50E
24.	VL 639	(VL 421 x CPAN 1535)
25.	VM 147	Inla-On-Inla-Bb/Y 50-Kal ³

One problem with the breeding for rust-resistance is that selection tests require high amounts of uredospores which have to be available at any time. Shortages in spore supply may arise, since the uredospores of many rust varieties are difficult to store at room temperature (Hassebrauk, 1970) and must furthermore be multiplied on living plants, which may cause problems during the winter months. Therefore, storing methods are necessary which are easy to apply and ensure long-term preservation of the spores. Most experiments on storage methods of uredospores from rust fungi described in the literature were made with cereal rusts. During the past years, rust-resistance has become increasingly important in grass breeding. According to Becker (1928) the storing ability varies among the rust species, and Bromfield (1967) found this also to be true for physiological races of *P. graminis* var. *tritici*. Thus we tested a number of storing methods for three grass rusts in extensive experiments. The following rusts were used: *P. coronata* var. *coronata*, *P. graminis* var. *poae* and *P. brachypodii* var. *poae-nemorialis*.

INTRODUCTION

We examined preservation methods for spores of different grass rusts of the species *Puccinia*. For the purpose of comparison, several preservation methods were applied to two cereal rusts, where they yielded slightly better results. Regarding the storage temperature, the life time of the spores increased with decreasing temperature. Preservation in liquid Nitrogen turned out to be the most favourable method. Deep-frozen spores received a heat treatment prior to further use. Vacuum dried spores maintained their viability less well over the storage period than stated in the literature. We put this down to the volume of the test jars, which was too large. A hydration of vacuum-dried spores improved the germination rate, but the results were unstable. The combination of both methods, namely the storage in vacuum at low temperature also proved to be favourable for preservation. We therefore presume that a heat treatment of dormant spores exerts a more positive influence on the germination ability than hydration, provided that the spores had been stored at a temperature <0°C before the treatment.

SUMMARY

Universität-GH Paderborn, Fachbereich Landbau, Labor für Biotechnologie und ökologische Phytomedizin, Windmühlenweg 25, D-4770 Soest, Germany.

BIRCKENSTADT, E.; HARDTKE, S.; LORENZ, E.; FAUL, V.H.

LONG-TERM PRESERVATION OF UREDOSPORES OF DIFFERENT SPECIES OF THE GENUS *PUCCINIA* ON GRAMINEAE

Cereal Rusts and Powdery Mildews Bulletin
Volume 20, Parts 1 & 2, 1992

In comparison with the above mentioned rusts, we also examined two cereal rusts: *P. coronata* var. *avenae* and *P. recondita* var. *tritici*.

MATERIALS AND METHODS

The uredospores were produced on the following plant species in the glasshouse: *P. coronata* var. *coronata* on *Lolium* sp., *P. coronata* var. *avenae* on oat, *P. graminis* var. *poae* and *P. brachypodii* var. *poae-nemorals* on Kentucky bluegrass, *P. recondita* var. *tritici* on wheat. The plants were inoculated with a spore-talcum mixture, which was dusted with a powder blower. They were then incubated for 24 hours in a humidity of 100%. About 10 to 14 days later we harvested the spores with a cyclone-collector following Cherry and Peet (1966). After harvesting, the spores were riddled and dried over silica gel for two days. After two days they were put into small jars (0,5-10 ml) in portions of ca. 20 mg and stored.

The applied storing methods fall into two groups: storage at low temperatures and storage under exclusion of oxygen (vacuumising). For both methods different temperatures and/or vacuumising periods were tested. Samples from each storage experiment were taken monthly and screened on water agar for germination. To compare the different rusts and methods with each other, the germinated spores from the samples were counted and the percentage calculated in relation to the original germination ability (prior to storage and/or treatment), which had been set to 100%. According to Mazur (1968), vacuum-dried spores of the genus *Puccinia* and *Uromyces* which have been stored below 0°C are not germinable in contrast to other fungus genera. They require a heat treatment. Having been taken out of the refrigerator, the frozen spores were therefore subjected to a heat treatment in a water bath of 45°C (Birckensstaedt, 1990). According to Rowell (1956), strongly dried uredospores of *P. graminis* var. *tritici* may be damaged, if they absorb the water too quickly, because an irregular swelling of the plasmalemma and the cell wall results in their separation from each other. Therefore Rowell (1984) recommends a slow hydration of the spores in a humid atmosphere. However, in the course of our experiments the results of the hydration varied considerably. We therefore did not apply to hydration to each sample of vacuum-dried stored spores but examined it separately.

RESULTS AND DISCUSSION

When evaluating our results and the literature, three questions have to be answered: what characterises a good storing method? Which results can be called satisfactory? What are the stored spores to be used for? In our own work (Birckensstaedt, 1990) with *P. coronata* var. *coronata* on *Lolium* sp. we found out that for experiments under more or less controlled conditions the spores should have a germination ability of 70-80%, in any case not lower than 50%. However, if the inoculation is carried out for spore production purposes only, even spores with very low germination rates (e.g. 10%) may be multiplied by repeated periods of leaf wetting.

All tested rusts maintained their germination ability the better, the lower the storing temperature used. There were differences among the

For *P. coronata* spp. storage in the refrigerator at +5°C for 2 months was unsuitable (Fig. 1). On the other hand, *P. recondita* var. *tritici* still maintained a germination ability of ca. 70% after a 6-month storage at +5°C (Fig. 1) as opposed to ca. 80% (Fig. 2) after the same period in the freezer at -30°C, where *P. coronata* var. *avenae* also stayed viable. At both temperatures, *P. graminis* var. *avenae* yielded average results. The germination ability decreased most strongly with *P. coronata* var. *avenae* (Fig. 1, 2). Stored in liquid nitrogen and in a special freezer at -80°C, however, all rusts maintained their original germination rate of 100% (Fig. 2).

Freeze-drying and vacuum-drying are used for the preservation of many microorganisms. Sharp and Smith (1952) applied both methods to *P. coronata* var. *avenae* and found vacuum-drying to yield the better results. When comparing vacuum-drying periods of different durations, we found out that the shortest period (5 min.) yielded the best results (Fig. 3). Figure 4 displays the reactions of four rusts after a vacuum-drying period of 2 hours. Once more *P. recondita* var. *tritici* proved to be the best and *P. coronata* var. *avenae* the most sensitive. *P. graminis* var. *avenae* again yielded average results, as did *P. brachypodii* var. *avenae-nemoralsis*. During the first two weeks after the vacuum treatment the strong decrease in the germination ability is typical especially with *P. brachypodii* var. *avenae-nemoralsis*. The comparison of the two subspecies of *P. coronata* in Figure 3 shows that the oat subspecies is less damaged by vacuum-drying and storage in vacuum than the grass subspecies of crown rust. On *P. graminis* var. *avenae* we finally examined how far the germination ability is affected by filling up the vacuum with an inert gas (Nitrogen). Figure 5 clearly illustrated that over the investigation period the germination rate was higher than a 2-hour vacuum-drying only (graph 2, vs. 1). It was even better when the 2-hour vacuum-drying was completely omitted and the air just replaced by Nitrogen. The graphs of the storing methods with vacuum-drying (without hydration) all display a similar trend: the germination ability decreases more or less strongly over a period of one year. However, it is conspicuous that during the first few weeks or months after the vacuum treatment the germination rates are still relatively high, although hydration had not been applied. Furthermore, the germination rates increase if the vacuum pressure is either reduced by shorter vacuum-drying periods (Fig. 3) or by releasing the vacuum with inert gas (Fig. 5).

Above we already mentioned that Rowell (1956) attributes the H₂O-damage to the separation of protoplast and cell wall. If this were the only reason, the germination rates in our experiments (Fig. 3, 4) should be expected to be low immediately after the vacuum treatment without subsequent hydration rather than only later. Presumably, as a result of strong vacuum pressure, the spores are not only damaged by shrivelling but also negatively affected by other processes. The water content for example sinks below a critical level, so that the loss of bound water causes irreversible damage and hydration can no longer yield the expected results (Mazur, 1968). This is certainly one reason for the unfavourable effect of long periods of vacuum-drying in our experiments, but perhaps also for the decrease of the germination rate with spores stored in vacuum. Over the time the 8-ml volume of the vacuum jars may have filled up with steam and/or further ingredients of the spores.

We conclude that methods for preservation of uredospores of rusts vary in their suitability. However, rust species and varieties have a different suitability for preservation which can be seen especially with the more favourable results of the cereal varieties, despite comparable conditions of spore multiplication. Thus the storage capability is genetically determined. This corresponds with the findings of Melching et al (1991) that there are always spores in a rust population which germinate without a heat treatment. For the storage of rust spores in vacuum we determined that the spores should fill the storage jar as high as possible.

Heat treatment of cold-dormant spores was a very reliable treatment. Bromfield (1964) reports success with the hydration of vacuum-dried spores. Sharp and Smith (1957) had already stated the same. However, they mention the unstable results of this special treatment, which corresponds to our own findings. Figure 6 shows a comparison of hydration and heat treatment. A quantity of spores was divided and was given to various treatments. The treatment exerts a considerably more positive influence on the germination rate than hydration. Figure 3 demonstrates the stimulating effect of a heat treatment on the spore germination clearly, in course of which germination capacities of a 100% resulted. According to Bromfield (1967) and Maheshwari and Sussman (1971) heat treatment and hydration with *P. graminis* var. *tritici* may partly be replaced by each other. This leads to the assumption that similar physiological processes are involved with both treatments. Mazur (1968) supposes that a strong dehydration may likewise induce dormancy. In their experiments Maheshwari and Sussman (1971) found some hints of how to explain the processes involved in cold-dormant spores. They report that with dormant spores not subjected to heat treatment the efflux of soluble and low-molecular substances in a buffer solution was higher than with heat-treated - that is to say, active - spores. This means that the membranes of dormant spores possess a higher permeability which can be removed by heat.

In the context of the discussion above, it is now clear why the following experiment led to good results. Vacuumised samples of *P. coronata* var. *coronata* were stored at -30°C and treated with heat prior to its use. Figure 7 shows only slightly decreasing rates of germination during 2, 5 years.

Depending on the rust species and on the aim of an application the following methods of preservation can be proposed:

1. The storage at temperatures > -30°C and subsequent heat treatment.
2. The storage of vacuumised samples at temperatures -30°C with subsequent heat treatment.
3. The storage in gaseous Nitrogen.

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- 1 = *Puccinia graminis*
var. *poae*
- 2 = *P. recondita*
var. *tritici*
- 3 = *P. coronata*
var. *coronata*
- 4 = *P. coronata*
var. *avenae*

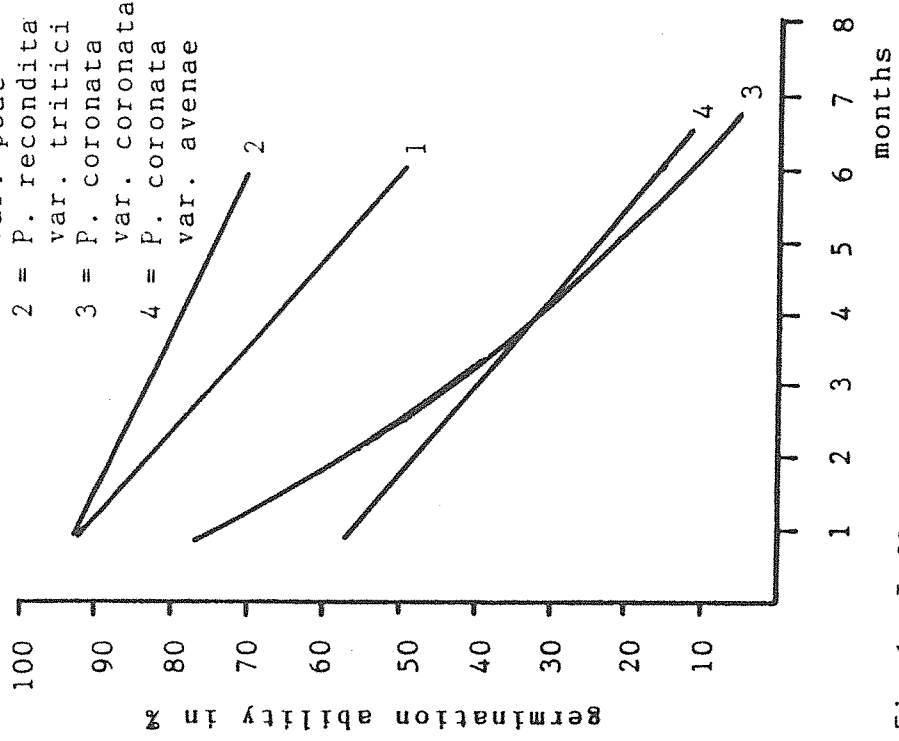


Fig.1: Influence of a storing temperature of +5°C on the germination ability of various rust varieties

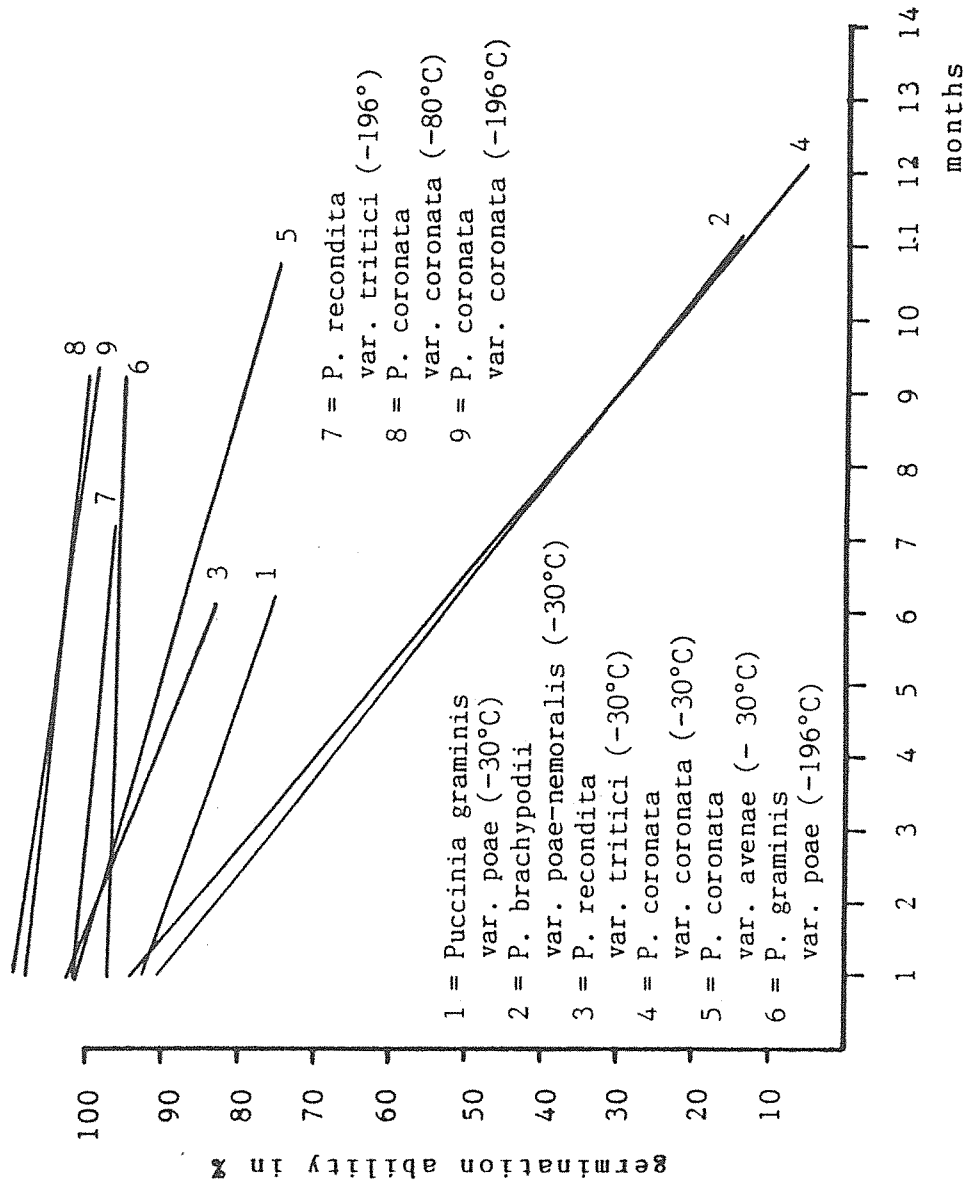


Fig.2: Influence of storing temperatures of -30°C, -80°C and -196°C with subsequent heat shock on the germination ability of various rust varieties

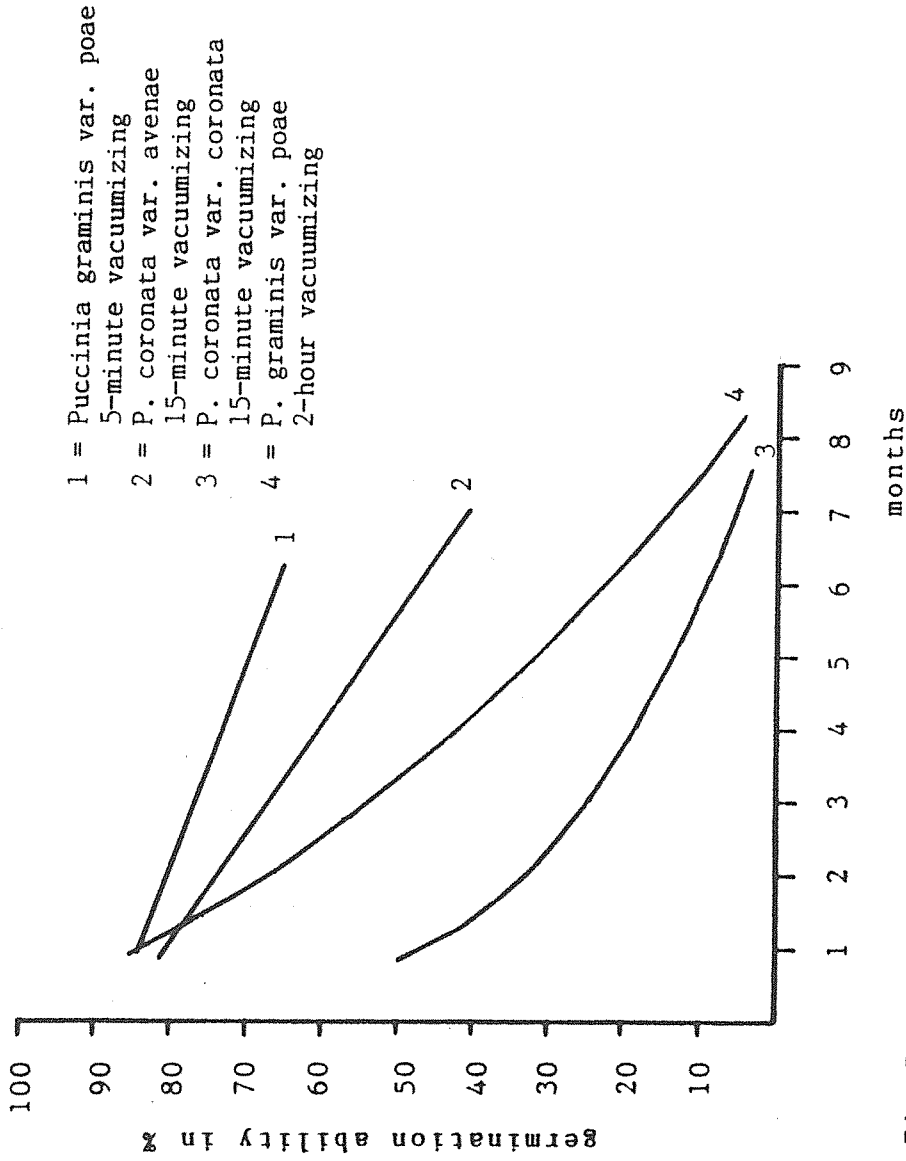


Fig.3: Influence of vacuumizing periods of different durations and subsequent storage in vacuum (at +5°C) on the germination ability of various rust varieties (without hydration)

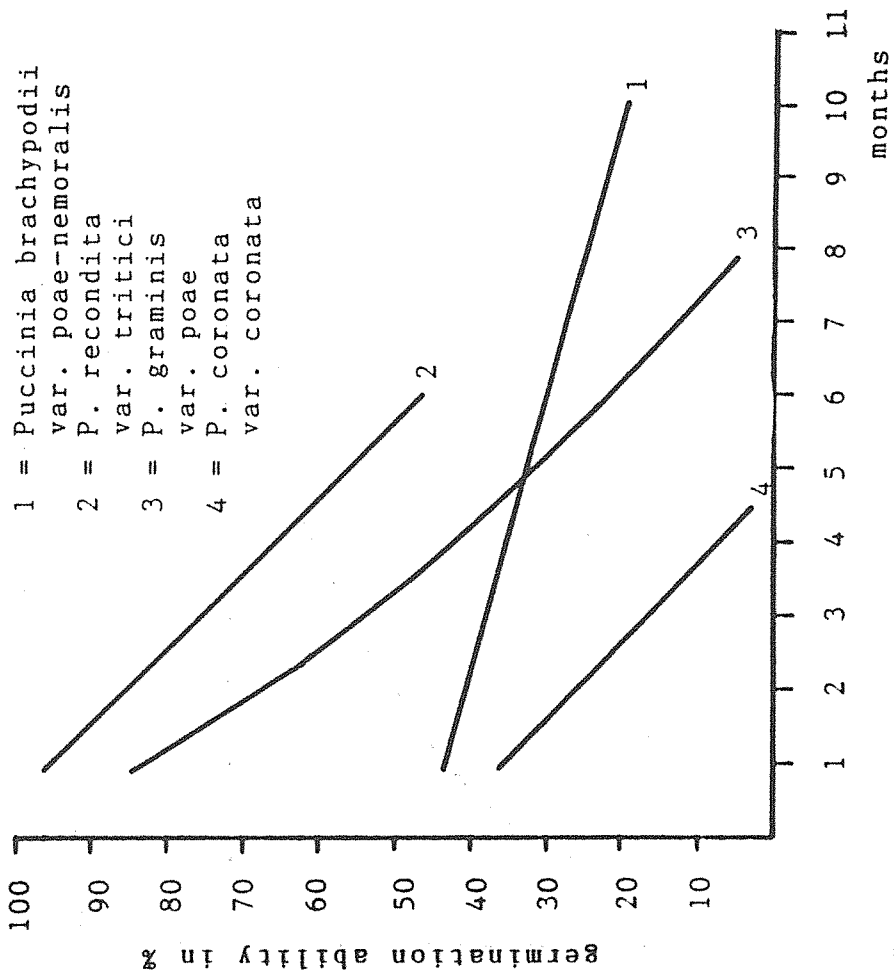


Fig.4: Influence of a 2-hour-vacuuminizing and subsequent storage in vacuum (at +5°C) on the germination ability of different rust varieties (without hydration)

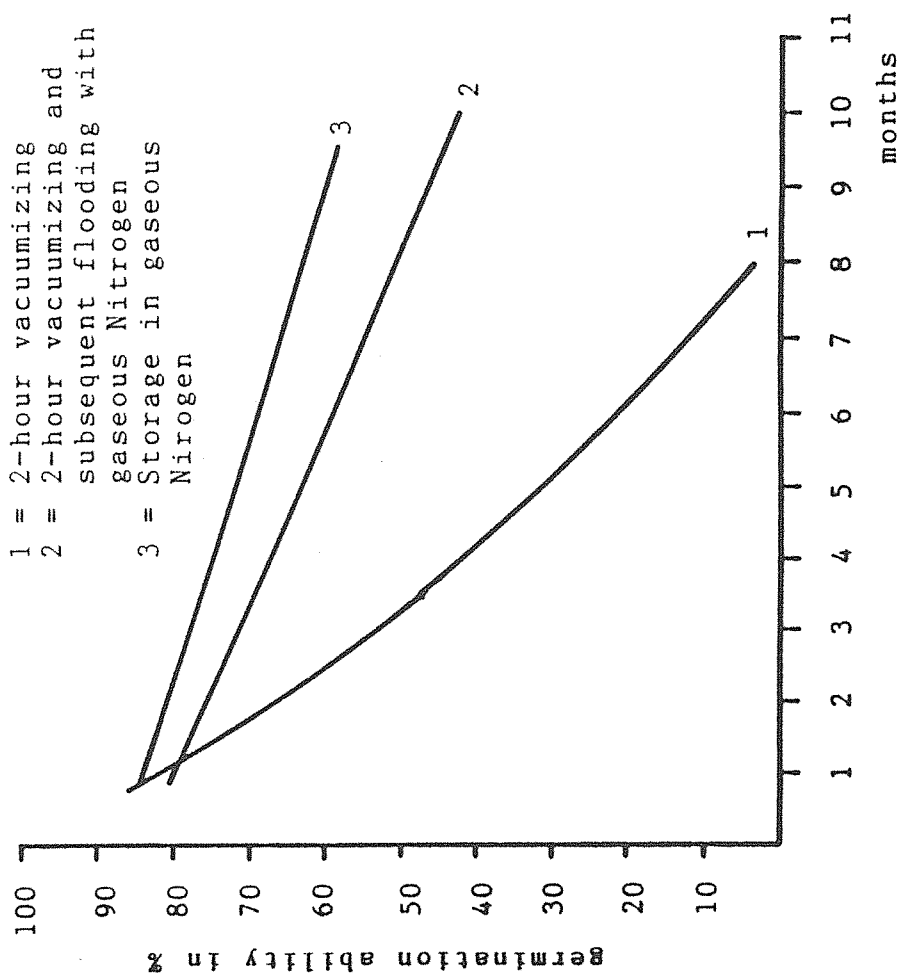
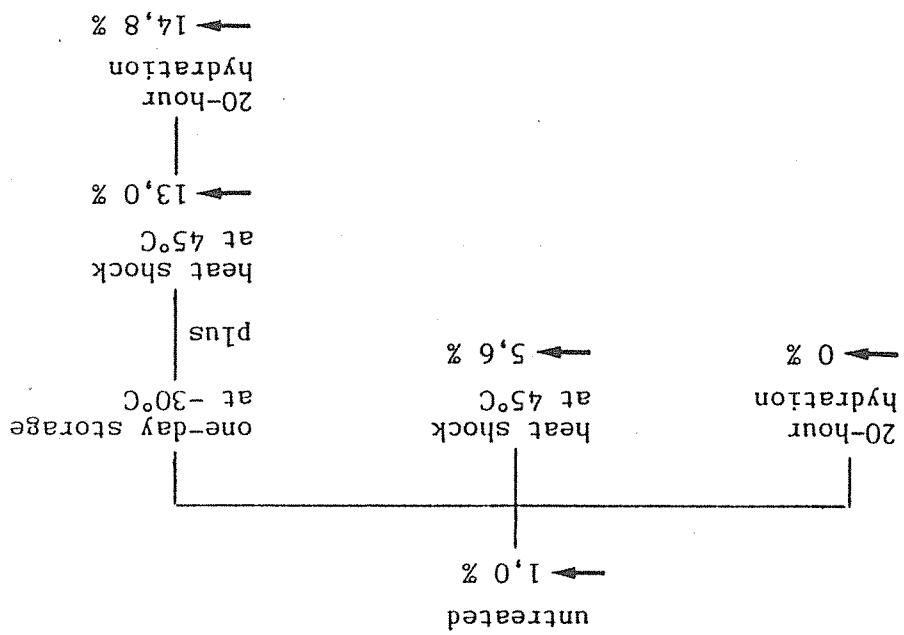


Fig.5: Influence of different storing methods in an oxygen-free atmosphere on the germination ability of Puccinia graminis var. poae (without hydration)

Fig. 6: Germination rates of a vacuum-dried spore sample of *Puccinia coronata* var. *coronata* stored for two years at +5° C after different special treatments.



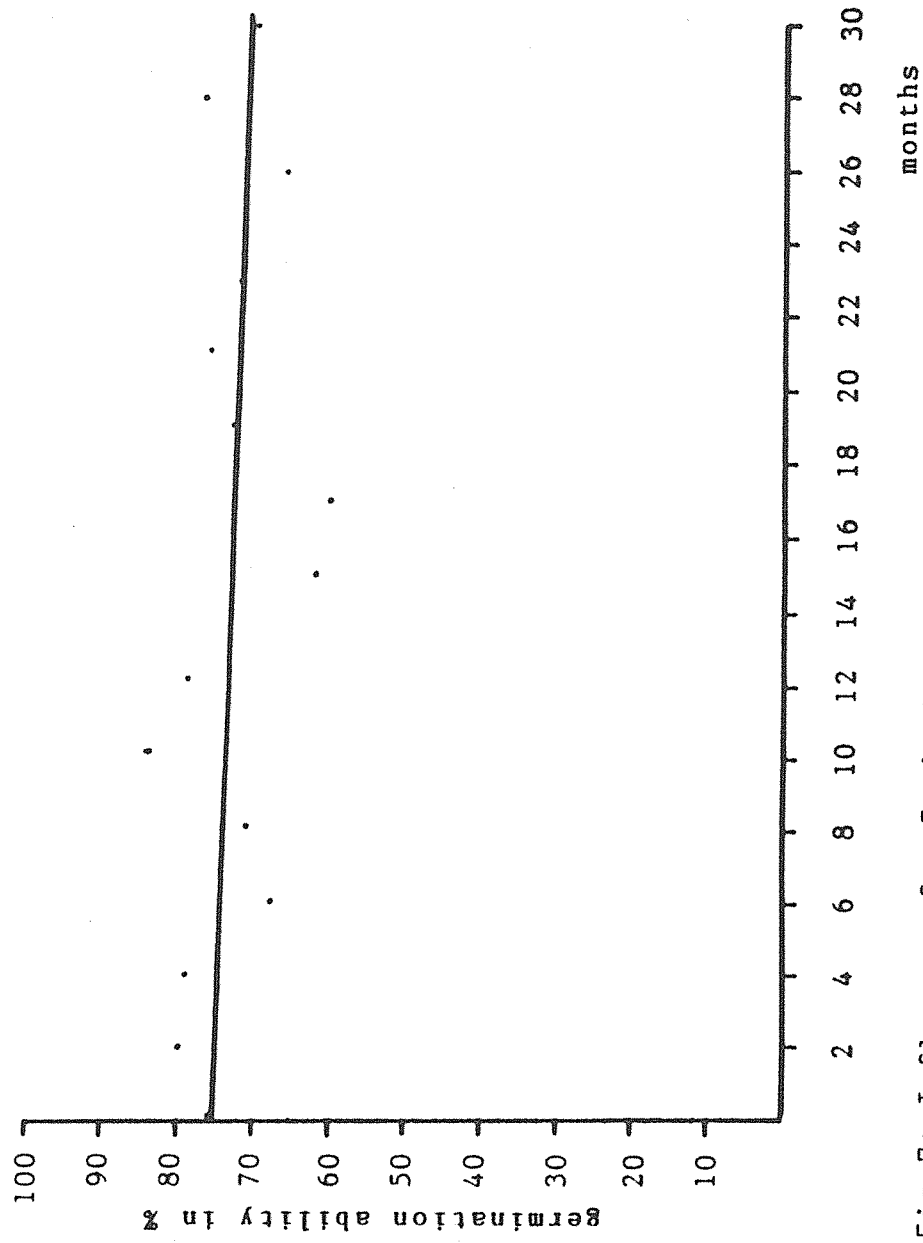


Fig.7: Influence of a 5-minute-vacuizing and subsequent storage at a temperature of -30°C of Puccinia coronata var. coronata (with subsequent heat shock)

NEW PATHOTYPES OF *Puccinia recondita* (BROWN RUST) IN THE UK AND THEIR
IDENTIFICATION IN THE FIELD AND CONTROLLED ENVIRONMENTS

S.D. ABDUL, A.J. TAYLOR AND T.W. HOLLINS

Plant Breeding International, Maris Lane, Trumpington, Cambridge
CB2 2LQ.

I. Present address: Cambridge Laboratory, John Innes Centre for Plant
Science Research, Norwich NR4 7UJ.

ABSTRACT

Seedlings and adult plants of thirteen UK local differential
cultivars along with cultivars Pastiche, Tara and Virtue were
evaluated for their response to new isolates of *Puccinia recondita*
plus a control pathotype, WBRP 85-31 in growth cabinets at 10, 15 and
25°C. The three new isolates, WBRP 90-10, WBRP 90-11 and WBRP 90-12
collected from infected Pastiche and Virtue crops were shown to differ
from each other as well as from the control pathotype. Isolates
WBRP 90-10 and WBRP 90-12 are shown to be new pathotypes. Isolates
WBRP 90-10 overcame Pastiche resistance at all growth stages and
equally at all temperatures. This was further confirmed by field
observations. Isolate WBRP 90-12 overcame Virtue resistance in the
field. Adult plant resistance(s) in cultivars Tara and Virtue were
shown to be less effective at high temperature (25°C) against isolate
WBRP 90-11 also collected from Virtue.

This demonstrated the importance of controlled environment in race
identification and genetic analysis of resistance of wheat to
P. recondita.

INTRODUCTION

Host resistance has been used with some success in the control of rust
pathogens. It has the advantage of having little or no direct cost to
the farmers. However, the specificity in the interaction between
host, pathogen and the environment leading to the expression of the
resistance or susceptibility require better understanding for the
effective deployment of resistance genes in agriculture.

The expression of resistance of wheat against brown rust (*Puccinia
recondita*) can change with temperature - a shift from susceptibility
to resistance may occur with increase in temperature or vice versa.
As early as 1926 Mains and Jackson noted differences in the resistance
of wheat cultivar 'Hussar' to some physiologic races of brown rust
after inoculation in autumn or winter compared with inoculation in
late spring. Rajaram *et al.* (1971) noted that at 24-27°C seedlings of
wheat cultivars W 3300 and W 3303 expressed resistance becoming
susceptible at 15-18°C, while the cultivar W 3301 changed from a

Pastiche, Tara and Virtue were field grown and received inoculum of either WBRP 90-10, WBRP 90-11 and WBRP 90-12 from a susceptible

Intermediate (I) and susceptible (S).
obtained were summarized into three categories: Resistant (R),
described by Stakman et al. (1962). For ease of presentation scores
temperature, the infection type was assessed using the system
expressed on the susceptible check cultivar Armada at a given
plants were inspected periodically and when symptoms were fully

by fluorescent tubes.
and 200µE/m²/s for seedlings and adult plants, respectively) supplied
set at 10, 15 and 25°C (+2°C) with 16 hours light (about 190µE/m²/s
isolates were grouped into three and transferred into growth cabinets
Seedlings as well as adult plants inoculated with each of the three
inoculation plants were incubated for 24 hours at 15°C (+2°C).
41-59, Zadoks et al., 1974) using a powder blower. Following
used to inoculate both seedlings and adult plants (growth stage
control pathotype WBRP 85-31 were separately dispersed in talc and
of two of the new isolates, WBRP 90-10 and WBRP 90-11 along with the
included as susceptible and resistant controls, respectively. Spores
growth cabinets. The cultivar Armada and line Lr 19 were also
along with the cultivars Pastiche, Virtue and Tara were evaluated in
Thirteen UK local differential cultivars (Clifford and Jones, 1984)

Three samples of *P. recondita*, WBRP 90-10, WBRP 90-11 and WBRP 90-12
were collected from wheat varieties Pastiche and Virtue crops in
Cambridge in July, 1990. These were subsequently increased on the
susceptible cultivar Armada and spores stored in liquid nitrogen.

MATERIALS AND METHODS

In 1990 the winter wheat varieties Pastiche, Tara and Virtue
previously known to be resistant to *P. recondita* in the field in the
UK were observed to be infected. Since the summer of that year was
characterized by relatively high atmospheric temperature (Table 1),
isolates obtained from these varieties were assessed to determine
whether infections were due to relatively high temperatures or as a
result of new virulence.

Reports on the sensitivity of some Thatcher backcross lines with
different brown rust resistance (Lr lines) were provided by Dyck and
Johnson (1983). Of these lines, Lr 18 was effective at low
temperature; Lr 16, Lr 17 and Lr 23 were effective at high temperature
and the groups Lr 2 and Lr 3 had stable resistance with some
increase in temperature.

resistant reaction at low temperature to a mesothetic reaction at high
temperature. Similarly the wheat cultivar Maris Fudrin expressed
temperature sensitive resistance that was effective only at high
temperature (Clifford et al., 1977; Hyde, 1982). In contrast, some
cultivars have been reported to be resistant at low temperature but
susceptible at high. The gene Lr 20 in the Thew background was
rendered ineffective at 30°C when inoculated with a pathotype
avirulent at 20°C (Jones and Deverall, 1977).

spreader variety (Armada) sown alongside each test plot. Diseased seedlings were transplanted into the spreader row in March. The cultivars Rendezvous and Apollo were used as resistant and susceptible controls, respectively. Disease score was taken as the percent leaf area infected.

RESULTS

Seedling reactions (Table 2) showed that all three isolates tested could be distinguished from each other with the UK set of differential cultivars based on their interactions with WBR 2, WBR 3 and WBR 4 differential groups. No isolate was compatible with the WBR 7 resistant cultivars at any temperature, although resistance was slightly decreased at low temperature with isolates WBR 85-31 and WBR 90-11.

Pastiche, Virtue and Tara all showed some seedling susceptibility to the standard isolate, WBR 85-31 and one of the two recently collected isolates, WBR 90-11. Hobbit seedlings showed a heterogeneous reaction to isolate WBR 90-10, particularly at 10° and 15°C where some plants were susceptible whilst others were resistant.

Cultivars in group WBR 2 all expressed seedling resistance against the standard isolate WBR 85-31 at 25°C only, although Hobbit had an intermediate reaction at 15°C also (Table 2).

Adult plants of Pastiche, Virtue and Tara were resistant in growth cabinets at all temperatures to isolate WBR 85-31 (Table 3). Pastiche was however, susceptible to the isolate WBR 90-10 across all temperatures (Table 3) and in the field (Table 4). This isolate also produced a heterogeneous response on adult plants of Hobbit (Table 3).

Virtue was susceptible to the isolate WBR 90-11 at 25°C and intermediate in response at 15°C (Table 3). Tara was intermediate at 25°C only. Both varieties were relatively resistant against this isolate in the field (Table 4). However, field plots of cultivar Virtue were susceptible to isolate WBR 90-12.

DISCUSSION

The three new isolates, WBR 90-10, WBR 90-11 and WBR 90-12 collected from Pastiche and Virtue were shown to differ from each other as well as from the standard pathotype WBR 85-31 based on their interactions with the differential cultivars and other cultivars as seedlings and/or adult plants in growth cabinets or in the field (Tables 2,3,4).

Isolate WBR 90-10, collected from Pastiche, was virulent on seedlings and adult plants of Pastiche at all temperatures used and isolate WBR 90-12 collected from Virtue, was particularly virulent on Virtue in the field. Both isolates represent new pathotypes and as they are found on large foci in seed multiplication plots in Cambridge area may be widespread, albeit as a small component, in the rust population.

The resistance of Virtue and to a lesser extent Tara adult plants to WBRP 90-11, was temperature sensitive and was only fully expressed at 10° and 15°C in controlled environment tests (Table 3), similar interaction has been reported by Clifford and Jones (1984). This could have resulted in the resistance of Tara and Virtue being ineffective in early summer of 1990 when unusually high temperatures were experienced (Table 1). However, field evaluation of these cultivars in 1991 showed them to be only slightly rusted (Table 4) presumably because the resistance was effective at the slightly lower summer temperatures experienced in that year.

Despite the heterogeneity observed among Hobbit seedlings, it is the only cultivar to show resistance to WBRP 90-10 from those in group WBR 2. Again adult plants of Hobbit and Fundin (WBR 2) expressed different responses to isolate WBRP 90-10 (Table 3). Some Hobbit plants were resistant to this isolate across temperatures, while others had compatible interaction. Fundin on the other hand had temperature sensitive resistance that was effective at 10°C, becoming ineffective with increase in temperature. Taking into account Fundin seedling resistance (effective at high temperature) against pathotype WBRP 85-31 (Table 2), this suggests the presence of additional resistance in this cultivar. Broder and Eversmeyer (1983) also demonstrated the presence of two types of resistances in Fundin - one effective at low temperature, and another effective at high.

The temperature sensitive resistance expressed by cultivars in group WBR 2 against isolate WBRP 85-31 may be due to Lr 13 and/or Lr 17, both effective at high temperature (Dyck and Johnson, 1983), as they have been reported in Hobbit (MacIntosh, pers. comm.). The resistance expressed against isolate WBRP 90-11 by cultivars Sappo, Sicco and Halberd in groups WBR 3 and WBR 4 could be due to the gene Lr 20. This gene has been identified in these cultivars (Dyck and Johnson, 1988).

Based on these observations it is recommended that genetic studies and race identification from disease surveys should be carried out under clearly defined environmental conditions as well as in the field to allow meaningful conclusions to be drawn.

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Table 1. Monthly average air temperature (°C) from 1987-1991 recorded at Plant Breeding International, Trumpington, Cambridge.

Month	1987		1988		1989		1990		1991	
	Max	Min	Max	Min	Max	Min	Max	Min	Max	Min
January	2.8	-1.8	8.3	4.0	9.1	3.4	9.9	3.8	7.0	1.5
February	6.5	-0.2	8.6	2.2	10.1	3.1	11.5	3.8	5.4	-1.8
March	-	-	10.0	3.5	12.2	7.8	13.3	4.0	12.7	4.7
April	15.2	5.8	13.4	0.2	10.7	3.0	14.1	2.5	12.9	4.2
May	14.8	5.5	16.7	8.4	20.1	7.2	19.5	6.5	14.7	6.7
June	17.3	9.1	18.6	9.5	21.8	9.3	19.1	10.1	16.6	8.8
July	20.8	11.1	20.1	11.5	24.8	13.0	23.6	11.4	23.6	13.1

Table 2. Summary of seedling infection types produced under different temperature (°C) regimes by *P. recondita* isolates WBPF 90-10, WBPF 90-11 and WBPF 85-31 on different wheat cultivars and lines.

Cultivar/	WBR	Lr ¹	WBPF ² 85-31	WBPF 90-10	WBPF 90-11	Line	group	gene	10°	15°	25°
Armada	S	S	S	S	S	S	S	S	S	S	S
Clement	(1)	26	S	S	S	S	S	S	S	S	S
Hobbit	(2)	17, 13	S	I	R	R	R/S	R/S	R-I	S	S
Fundin	(2)		S	R	R	R	S	S	S	S	S
Norman	(2)		S	S	R	R	S	S	S	S	S
Sappo	(3)	20	S	S	S	S	S	S	S	S	S
Sicco	(3)	20	S	S	S	S	S	S	S	S	S
Halbred	(4)	20	S	S	S	S	S	S	S	S	R-I
Brigand	(5)	13	S	S	S	S	S	S	S	S	S
Huntsman	(5)	13	S	S	S	S	S	S	S	S	S
Gamin	(6)		S	S	S	S	S	S	S	S	S
Sterna	(7)		I-R	R	R	R	R	R	I	R	R
Sabre	(7)		I-S	R	R	R	R	R	I	R	R
Pastiche			I	S	S	S	S	S	S	S	S
Tara			S	S	S	S	S	S	S	S	S
Virtue	13		S	S	S	S-I	R	I	I-S	R	R
Lr 19	19		R	R	R	R	R	R	R	R	R

1 Lr gene reported by the following workers:

Lr 26 (R. Johnson pers. comm.)
 Lr 13, 17 (R.A. McIntosh pers. comm.)
 Lr 20 (Dyck and Johnson, 1988).

2 WBRP = Wheat Brown Rust Pathotype

Table 3. Summary of adult plant infection types produced under different temperature ($^{\circ}\text{C}$) regimes by *P. recondita* isolates WBRP 90-10, WBRP 90-11 and WBRP 85-31 on different wheat cultivars and lines.

Cultivar/ WBR	Lr	WBRP 85-31	WBRP 90-10	WBRP 90-11	group	gene	10°	15°	25°	10°	15°	25°	10°	15°	25°
Armada		S	S	S	S	S									
Lr 19		R	R	R	R	R									
Hobbit	(2)	R	R	R/S	R/S	R/S									
Fundin	(2)	I	R	I-S	S	S									
Brigand	(5)	R	R	R	R	S									
Pastiche		R	R	S	S	S									
Tara		R	R	R	R	R									
Virtue		R	R	R	R	R									

Table 4. Percent leaf area infected with brown rust 16th July 1991 (field data).

Cultivar/WBRP isolate	85-31	90-10	90-11	90-12
Pastiche	30*	50	13	8
Tara	1	0	0	1
Virtue	15	8	10	55
Rendezvous	0	1	1	2
Brigand	33	10	26	53
Apollo	60	55	68	43

* Probably contaminated plot.

These are coded from 1-10 and are placed in a fixed linear order in groups of three numbering from the right; new resistant genotypes are thus added to the left.

Code	Genotype	C.I. Number	Gene	Reference
1	Sudan	6489	Pa	Roane and Starling (1967)
2	Peruvian	935	Pa ₂	Starling (1956)
3	Ribart	-	Pa ₃	Bruckner (1971)
4	Gold	1145	Pa ₄	Roane and Starling (1967)
5	Quinn	1024	Pa ₅ (+Pa ₂)	Roane and Starling (1967)
6	Bolivia	1257	Pa ₆ (+Pa ₂)	Roane and Starling (1970)
7	Cebada Capa	6193	Pa ₇	Parlevliet (1976)
8	Egypt 4	6481	Pa ₈	Tan (1977a)
9	-	C.I. 1243	Pa ₁₀	Clifford and Jones (1981)

Table 1: Differential barley cultivars used in the UKCPVS of *P. hordei*

Central to formal virulence analysis is a set of differential host cultivars. A set of eight was assembled based on published and unpublished genetic information on their resistance (Pa) genes (Clifford, 1974). Since then two additional cultivars, namely C.I. 1243 (Pa₉) and the former East German cv. Triumph (Pa₁₀) have been added giving the current set of ten (Table 1).

MATERIALS AND METHODS

In 1976, a meeting was held at the 4th European and Mediterranean Cereal Rusts Conference in Interlaken, Switzerland at which the author presented a proposal concerning the choice of differential host genotypes and technical procedures for monitoring virulence genes in *Puccinia hordei*. The results were published in the Cereal Rust Bulletin (Clifford, 1977). At the time, the author proposed the adoption of the octal/binary system (Gilmour, 1973) for race nomenclature as it met the requirements of a formal system of nomenclature. This paper reports the application of the system to virulence analysis of *P. hordei* populations conducted under the UK Cereal Pathogen Virulence Survey (UKCPVS).

INTRODUCTION

AFRC Institute of Grassland and Environmental Research, Welsh Plant Breeding Station, Plas Gogerddan, Aberystwyth, Dyfed SY23 3EB, UK.

B.C. Clifford

APPLICATION OF THE OCTAL/BINARY NOTATION SYSTEM TO VIRULENCE NOMENCLATURE IN *Puccinia hordei*, THE CAUSE OF BROWN RUST OF BARLEY

Following inoculation and incubation under standard conditions (Clifford, 1977), the host responses are then assessed according to standard reactions (Table 2) and classified bimodally as either resistant (R) or susceptible (S).

Table 2. Reaction classes employed in the *F. hordei*: barley system

Class	Reaction	Sporulation	Host Response
R	Of	None	None
R	Oc	None	Chlorotic fleck
R	On	None	Necrotic fleck
R	1	Slight	Necrosis
R	2	Moderate	Necrosis
S	3	Moderate	Chlorosis
S	4	Heavy	Slight chlorosis

NB Mixed reactions may occur on the same leaf

To obtain the unique octal notation for each virulence combination possible, the reaction classes are first assigned a binary number where R = 0 and S = 1. As the differential cultivars are assigned a fixed linear order and are grouped into sets of three, the corresponding binary triplets can then be assigned their correspondence octal number.

Binary Triplet Octal Number

000	0
001	1
010	2
011	3
100	4
101	5
110	6
111	7

The virulence combination or race designation is a series of octal numbers which, in the case of *F. hordei* where there are 10 differentials, has up to 4 digits.

For example, Race Octal 615:

Differential:	10	9	8	7	6	5	4	3	2	1	0
Reaction:	1	3	4	On	2	2	4	R	R	0c	4
Class:	R	S	S	R	R	R	S	S	R	S	S
Binary:	0	1	1	0	0	0	0	1	1	0	0
Octal:	0	6	1	0	1	1	5	1	0	1	1

RESULTS

In 1979, the octal system was introduced and applied to races of *P. hordei* previously identified in the UKCPVS (Jones and Clifford, 1980) (Table 3).

Table 3. Octal/binary system for the designation of virulence gene combinations carried by specific races of *Puccinia hordei*.
Oth.

Race	Differential Number									Octal Number	Year Identified	
	1	2	3	4	5	6	7	8	9			
A	0	0	0	0	0	0	0	0	0	0	11	1968
B	1	0	0	0	0	0	0	0	0	0	211	1968
C	1	0	0	0	0	0	0	0	0	0	212	1968
D	0	0	0	0	0	0	0	0	0	0	32	1968
E	1	0	0	0	0	0	0	0	0	0	271	1968
F	1	0	0	0	0	0	0	0	0	0	273	1969
G	0	0	0	0	0	0	0	0	0	0	10	1069
H	0	0	0	0	0	0	0	0	0	0	70	1969
J	1	0	0	0	0	0	0	0	0	0	232	1969
K	1	0	0	0	0	0	0	0	0	0	251	1973
L	1	0	0	0	0	0	0	0	0	0	210	1973
76/2	0	1	0	0	0	0	0	0	0	0	257	1976
76/3	1	1	0	0	0	0	0	0	0	0	673	1976
76/12	1	1	1	1	1	1	1	1	1	1	677	1976

* R = 0 S = 1

At that time, nine differential cultivars were employed following the addition of C.I. 1243 (Code No. 9) in 1976. This genotype carries gene Pa₉ (Tan, 1977b) which was also suggested to be present in the popular cv. Triumph (Waltur and Lehmann, 1980). However, evidence from virulence analysis (Clifford and Jones, 1981) indicates that cv. Triumph carries an additional factor and this has been designated Pa₁₀. The genotype was incorporated into the standard differential set, at No. 10 in 1985 (Clifford and Jones, 1986). Up to the present time, a range of different virulence combinations (races) has been identified with complex combinations occurring with high frequency. The results for the last five years data that are available are summarised in Table 4.

Table 4. Frequencies of *P. hordei* virulence combinations identified in the UKCPVS 1987-1991.

Octal Designation	BRV* Factors	Frequency
1673	1,2,4,5,6,8,9,10	0.54
1653	1,2,4,6,8,9,10	0.30
673	1,2,4,5,6,8,9	0.12
1657	1,2,3,4,6,8,9,10	0.04
1677	1,2,3,4,5,6,8,9,10	0
Number of Isolates		
		46
		60
		73
		49
		53

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ACKNOWLEDGEMENTS

The UKCPVS for barley brown rust is conducted at IGER by E.R.L. Jones and is funded by the UK Home-grown Cereals Authority.

Small plot trials were conducted in Prague-Ruzyne with winter wheat Danubia infected by yellow rust race Clement. The size of the plot was 2m². There were four replications. Five plants were infected (the first leaves) by urediniospores of *P. striiformis* followed by dewing and closing under a glass cylinder for 3 days. The infection was disseminated from these infected plants onto other plants. Disease severity was evaluated as the leaf area percentage of each leaf affected by yellow rust on 40 culms per replication on a 9 points scale (9 = 0%, 1 = 96-100%, Coakley et al.; 1983). Disease severity was expressed as cumulative percentage of incidence during the whole

MATERIALS AND METHODS

The purpose of this study was to find out the influence of these weather factors on the overall yellow rust severity in winter wheat.

The problem of yellow rust in Czechoslovakia has been practically solved in the last 20 years by breeding for resistance and by growing resistant cultivars. In Europe, however, yellow rust remains an important disease (Bartos et al.; 1991). Temperature, precipitation and sunshine rank among those factors which influence the rise and course of disease epidemics.

INTRODUCTION

In small plot trials the influence of temperature, precipitation and sunshine (the beginning of epidemic and the whole period of the epidemic) on the overall yellow rust (race Clement) severity in winter wheat (Danubia) were studied. Correlation coefficients indicate a positive, highly significant correlation between the overall disease severity and the sum of precipitations, the sum of average daily temperature, the length of incubation period, number of days with maximal temperature under 5°C and number of days with average temperature between 5 and 10°C in the initial period of epidemic development. A high negative correlation between the whole disease incidence and the sum of average daily temperature supports the importance of colder weather for the development of yellow rust epidemic.

SUMMARY

Research Institute for Crop Production, Prague

VECHET L.

F. SP. TRITICI

INFLUENCE OF SOME WEATHER FACTORS ON EPIDEMIC OF Puccinia striiformis

Cereal Rusts and
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Vol. 20 parts 1 & 2 1992

The incubation period was the longest in 1990 when the sum of average daily temperatures was also highest. Gopalan and Manners (1984) ascertained that germination of *P. striiformis* was better when spores were formed in cold and wet conditions than when weather was dry and warm. McGregor and Manners (1983) mentioned that increasing of temperature between 7-20°C shortened the latent period and restricted the age of sporulating leaves. Also production of spores reached its peak earlier and fell faster with increasing of temperature between 7 and 20°C. There was a medium strong positive dependence of the total disease severity on the number of days with minimal temperature below

the beginning of the epidemic is obviously related to the length of the incubation period. Positive correlation of disease severity to the sum of temperatures in sum of precipitations (k) during the first incubation period. Occurrence of disease was also the lowest. There was also the lowest trials the lowest number of rainy days (m) was in 1989, when the infection to appear, leaves must be wet for at least 6 hours. In our example Shaner and Powelson (1971) indicate that for the yellow rust water and temperature for successful infection by *P. striiformis*. For incubation period by 83.4%. A number of authors mention importance of 99.2%, the sum of daily temperatures by 95.8% and the length of incubation period influenced the total disease incidence of plant by determination showed that the sum of precipitations in the first development (the first incubation period). Calculated coefficient of temperature between 5 and 10°C (i) in the initial period of epidemic with maximal temperature under 5°C (h) and number of days with average temperature (d), the length of incubation period (e), number of days (a) and the sum, of precipitations (k), the sum of average daily experiments (Tab.1a). Correlation coefficients indicate a positive, highly significant correlation between the overall disease severity of yellow rust fluctuated in different years of the

RESULTS AND DISCUSSION

Factors of time were the length of the first incubation period of *P. striiformis* (since artificial infection), the beginning of epidemic and the whole period of the epidemic. The extent of disease severity was expressed by the area under the curve of disease progress function: $\sum_{i=0}^n f(x_i) \cdot (x_i - x_{i-1})$, where $f(x)$ is value of discontinuous function (percentage of disease severity) and x is time. Linear regressions were used to express the relationship, between the disease incidence and the influence of selected weather factors. Weather influence was expressed by correlation coefficient. For assessing the suitability of the equation, coefficient of determination was used.

14 days preceding by 7 days the date of observation. Values in two variables: 14 days preceding the date of observation and consideration. Sets of meteorological data were used as independent average daily sunshine and sum of sunshine were taken into temperature, average daily precipitation, sum of the precipitation, average daily temperatures, number of days with minimal and maximal period of the epidemic, dependent value. From weather factors, average daily temperature (min t + max t), sum of average daily

The results indicate that the most important period for the relevance of epidemic due to *P. striiformis* is the beginning of the epidemic, the first incubation period. Favourable weather in this period, high sum of precipitations, higher sum of average daily temperatures together with longer first incubation period, with lower sum of temperatures during the whole period of epidemic help to initiate the epidemic and consequently to increase the incidence of yellow rust in wheat.

0.0°C (j), on the sum of sunshine (n), on average daily rainfall (l) in the initial period of epidemic development, on number of days with average temperature between 5 and 10°C (H) and on days with average temperature between 7 and 12°C (I) during the whole period of epidemic (Tab.2). A negative, medium strong dependence of the overall occurrence of the epidemic on average daily temperature (e), on number of days with average daily temperature above 10°C (g) and on number of days with minimal temperature above 15°C (f) supports the advantage of a colder initial period for the development of epidemic. Also a high negative correlation between the whole disease incidence (a) and the sum of average daily temperatures (c) for the whole period of epidemic supports this deduction. Similarly, a negative, medium high dependence on number of days with average daily temperature (D), on number of days with average temperature above 10°C (E) and with temperature above 20°C (G) during the whole period of epidemic supports this assumption. McGregor and Manners (1988) stated optimal temperature for an increase and sporulation of *P. striiformis* to be 9 to 16°C. Sharp (1965) considers the optimal temperature for germination and infection by yellow rust 7°C during most of the period. There was no correlation between the extent of the whole disease incidence (a) and the average time of sunshine (o) for the first incubation period, the whole epidemic period (B) and the sum of sunshine (J), average period of sunshine (K), the sum of precipitations (L) and average daily precipitation (M). It is probable that sunshine can influence yellow rust together with temperature. A similar phenomenon was recorded in *Erysiphe graminis* f. sp. hordei (Vechet et Kocourek; 1986).

Table 1. Disease severity (cumulative percentage of incidence) in winter wheat (Danubia) affected by yellow rust (the race Clement) in field conditions, a - area under the curve. The first incubation period of *P. striiformis* (period after artificial infection) : b - date of infection, c - length of incubation period, D - sum of average daily temperature, e - average daily temperature, f - number of days with maximum temperature above 15°C, g - number of days with average daily temperature above 10°C, h - number of days with minimal temperature under 5°C, i - number of days with average daily temperature 5-10°C, j - number of days with minimal temperature under 10°C, k - sum of precipitations, l - average daily precipitation, m - number of days with precipitation, n - sum of sunshine, o - average daily time of sunshine, r - coefficient of correlation, R² - coefficient of determination. Ruzhne.

	1988	1989	1990	1991	r	R ²
a	1451.40	430.60	5531.84	1796.04		
b	20.4	11.4	10.4	6.5		
c	14	15	22	18	0.913	0.834
d	169.65	162.80	197.10	165.10	0.977	0.958
e	12.13	10.85	8.96	9.17	-0.633	0.401
f	10	9	6	4	-0.435	0.189
g	10	9	5	3	-0.486	0.236
h	8	11	16	13	0.785	0.623
i	3	6	17	15	0.737	0.543
j	4	1	4	1	0.616	0.379
k	14.5	9.1	54.9	18.8	0.996	0.992
l	7.27	1.52	6.60	2.69	0.419	0.176
m	13	6	10	7	0.306	0.094
n	121.2	89.8	145.2	164.6	0.530	0.281
o	8.66	5.61	6.60	9.14	-0.106	0.011

Table 2. Disease severity (cumulative percentage of incidence) in winter wheat (Danubia) affected by yellow rust (the race Clement) in field conditions, a - area under the curve. The total period of epidemic: B - period of epidemic, C - sum of average daily temperatures, D - average daily temperature, E - number of days with average temperature above 10°C, F - number of days with average temperature 10-20°C, G - number of days with temperature above 20°C, H - number of days with average daily temperature 5-10°C, I - number of days with average temperature 7-12°C, J - sum of sunshine, K - average daily sunshine, L - sum of precipitations, M - average daily precipitations, N - number of days with precipitations, r - coefficient of correlation, R² coefficient of determination. Kuznye.

	1988	1989	1990	1991	r	R ²
a	1451.40	430.60	5531.84	1796.04	-0.238	0.057
B	68	70	69	70	-0.841	0.707
C	937.8	872.5	826.4	569.2	-0.509	0.259
D	13.79	12.46	11.98	12.42	-0.381	0.145
E	59	70	53	41	-0.556	0.309
F	50	67	52	36	-0.332	0.110
G	0	3	1	5	0.641	0.411
H	6	11	23	23	0.560	0.314
I	8	19	23	16	0.239	0.057
J	383.8	613.6	589.8	521.5	0.175	0.031
K	4.9	7.2	7.7	9.7	0.108	0.012
L	231.47	94.1	109.7	66.5	-0.103	0.011
M	3.40	1.34	1.59	0.95	-0.336	0.126
N	38	32	26	20		

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Twenty-seven cultivars from the National Genetic Stock Nursery and eight wild species of wheat were sown in the field. Each entry was represented by one row of one metre length. The inoculum of a mixture of races/biotypes of stem rust was sprayed on 60 days old plants when the field was sufficiently wet to create an artificial epiphytotic of rust. The rust infection types were recorded at a late stage by using modified Cobb's Scale (Peterson et al., 1948). The average temperatures in the field during crop growth ranged between 8.9°C to 24.7°C with 62 per cent relative humidity.

MATERIALS AND METHODS

Genetic variability for resistance is essential for the success of breeding rust resistant varieties. The information on the interaction between the specific genotypes of the host and races/biotypes of pathogen is the basis for utilizing diverse sources of resistance. Attempts were made to study the host parasite interaction in wheat genetic stock and stem rust races/biotypes to identify sources of resistance.

Rust reduces vigour, seed-filling and root growth and thereby yield by increasing transpiration, respiration and reducing photosynthesis in the plant. Control of stem rust is quite difficult by use of chemicals and cultural practices for which resistant varieties are the best and cheapest means. Hence, sources of resistance are the pre-requisite of breeding for rust resistance.

INTRODUCTION

Twenty seven cultivars and eight wild species of wheat were tested by inoculating individual races/biotypes of stem rust at the seedling stage and at the adult plant stage in the field by creating an artificial epiphytotic of mixture of races of stem rust. The cultivars CPAN 1955, HD 2285, HD 2315, HS 207, HW 741, VL 490 and WG 2109 were resistant in both seedling and adult plant stages. The wild species Triticum carthlicum and Triticum dicoccoides of wheat were resistant in adult plant stages only.

SUMMARY

Regional Wheat Rust Research Station, Mahabaleshwar-412 806 (India)

R.T. Sapkal, R.J. Patil and J.V. Patil

HOST PARASITE INTERACTION WITH SPECIAL REFERENCE TO STEM RUST OF WHEAT

Cereal Rusts and
Powdery Mildews Bulletin

Vol. 20, Parts 1 & 2, 1992

Seven day old seedlings of all the entries raised in four inch diameter earthen pots were inoculated with the inoculum of individual races/biotypes of stem rust. After inoculation the plants were exposed to high humidity in a moist chamber for 24 hours and then transferred to a glasshouse bench for disease development. The average temperatures during seedling testing period ranged between 17°C to 36.2°C with relative humidity of 67 per cent in the glasshouse. Observations regarding pustule types were recorded using the key suggested by Stakman and Levine (1922).

RESULTS AND DISCUSSION

The seedling and adult plant reactions are presented in Table 1. The percentages of the cultivars under study are listed in Table 2.

The data in respect of seedling and adult plant reactions of the cultivars tested revealed that the cultivars CPAN 1955, HD 2285, HD 2315, HS 207, MW 741, VL 490 and WG 2109 were resistant to all the races/biotypes at seedling as well as adult plant stages. The cultivars CPAN 1909, CPAN 1956, HD 2190, MH 840 and MW 147 although susceptible to a few races/biotypes in seedling stage exhibited adult plant resistance. The cultivars BW 11, HD 2009, HD 2329, HI 1077, showed resistance to all the races at the seedling stage but were susceptible at the adult plant stage. This indicates that cultivars having resistance at seedling stages to all races/biotypes do not necessarily possess adult plant resistance. This observation is in confirmation of the cultivars HD 2190, HD 2315, MH 741, MH 840, VL 490 and WG 2109 for 4 to 5 years as reported by Agarwal (1986). These cultivars will be useful as parents in breeding programme.

None of the wild species tested against individual races/biotypes showed resistance against all the races/biotypes of stem rust at the seedling stage. However, two wild species, viz. *Triticum carthillicum* and *Triticum dicoccoides* showed resistance at the adult plant stage.

ACKNOWLEDGEMENT

Authors are thankful to Dr J P Tandon, Project Director (Wheat), I.A.R.I., New Delhi for supply of seed of the cultivars studied.

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Table 1 : Seedling and adult plant reactions of the wheat cultivars tested against stem rust races/biotypes

Cultivar	R14	15	17	21A	21A1	34	40	40A	42	42B	42B2	117A	117A1	122	184	295	Field reactions
1. BW 11	0;	0	0	-	0	0;-1	0	2+	0;	0	2	2	2	2	0;	1	40's
2. CPAN 1909	1	1	1	-	3	0	1	1	1	0	0	1	1	1	2	1	20 MR
3. CPAN 1955	1	1	0	-	1	0	0	1-2	0;	2	0	0	0	2	1	1	20 MR
4. CPAN 1956	2	1-2	1	-	3	3	2	1	3	1-2	2	0	0	3	3	1	10 MR
5. DWL 5023	2+	0	4	4	2	2	0;-1	3	0	2	4	3	4	2	3	4	40's
6. HD 2009	0	-	1	0	0	0	0	2	1	0	0	0	1	1	1	1	60's
7. HD 2190	1	-	0;-1	1	0	0	1	1	2	-	1	2	0;	0	3	2	30 MR
8. HD 2270	0	0	0	0	0	0	0	0	2	2	0;	0;-1	0;-1	2	0;-1	2	40 MS-TS
9. HD 2285	0	0	2	2	0	1	0;-1	2+	0	0	1	1	1	2	0;-1	2	20 MR
10. HD 2315	1	0	-	-	0;-1	1	1	0	-	-	0	-	0	2	1	1	30 MR
11. HD 2329	1	0	0	1-2	0	0	0;	2	2	0	0;	1	1	2	0;-1	1	40's
12. HD 2428	2	1	2	2	1	1-2	0;	3	2	0	2	2	1-2	2	0;	3	50 MS
13. HI 784	0	2	0	2	0;	2	0	3	0	0	0;	2+	2	3	3	3-4	40 MS
14. HI 1077	0;	1	0;-1	2	0;	0	0;-1	2	1-2	0	0	2	2	2	1	2	30's
15. HS 207	0	0;	0	0	0	2	0;	0	0	0	0	0	0	2	0	1	30 MR
16. HW 741	1	-	0;	0;	0	0	0	1	0;	0	1	1	1	1	0;	1	20 MR
17. HW 840	0	3-4	-	-	0	4	4	4	-	-	0	-	0	0	0	3	10 MR
18. JU-12	0;	0;-1	2	2+	2+	1	0;	2	0;-1	2	3	3	3	3	2	3	30's
19. K 7410	1	0	2+	1	2	2+	0;	4	2	2	2	2	2	3	0;-1	3	40's
20. K 8020	1	1	2+	0	2+	1	0;	0	2	0	3	2	3	3	2+	2	60 MS

Table 1 : Seedling and adult plant reactions of the wheat cultivars tested against stem rust races/biotypes continued

Cultivar	R14	15	17	21A	21A1	34	40	40A	42	42B	42B2	117A	117A1	122	184	295	Field reactions	
1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19
21. PBW 154	0	0;-1	2	2	0	1	0;-1	0;	3	2	2	2	2	1	2	0;-1	2	60 MS
22. PBW 175	1	0;	2	2	2+	1	2+	2	2+	2	0	1	1-2	2	2	0;-1	1-2	40 MS
23. VL 490	0	2	-	-	-	1	2	2	2	-	-	2	-	1-2	2	0	1	10 MR
24. VL 616	0	0	2	2	3	0	-	0	3	2	0	0;	2	3	2+	3	3	30 MS
25. WG 2109	1	0	0	0	-	0	0;	2	0;-1	1	0	1	0	1	0;	0	1	10 MR
26. WH 147	1+	2	2	2	1	4	2	0;-1	0	2	0	3	2	4	2	2+	3	20 MR
27. WH 416	0	2	0	0	0	0	1	1	0	0	0	0	0	0	3	0	3	60's
28. T.carthilicum	3	0;-2	3	3	3	0	4	3	3-4	0	0	3	4	3	3-4	3	4	20 MR
29. T.turgidum	0	0;	1	1	4	0	2	0	4	0;	0	0;	3	3	1-2	4	0;	40's
30. T.timopheevi	3	4	4	4	4	4	4	4	3	0	0	4	-	3	4	4	4	50's
31. T.sphaerococcum	3	4	4	4	4	4	4	4	4	0	0	4	4	3	4	4	4	50's
32. T.compactum	3	4	4	4	4	4	4	4	4	4	0	4	4	3	3	4	4	50's
33. T.persicum	4	0;-1	4	4	4	0	4	3	2	0	0	4	4	4	4	4	4	50 MS
34. T.dicoccoides	2	0	2	2	3	2	1	0;	2	0	4	2	0;	4	3	3	3	40 MR
35. T.pyramidale	4	3	3	3	4	0	3-4	3	4	4	4	3	3	4	4	3-4	4	TS

R = Resistant, MR = Moderately Resistant, MS = Moderately Susceptible, S = Susceptible, T = Trace

0 - Immune, 0, - Very Resistant, 1- Resistant, 2- Moderately Resistant, 3- Moderately Susceptible, 4- Susceptible

Table 2. Percentage of the cultivars tested against Stem Rust of wheat

1.	BW 11	KVZ-Tanori-71 x Tito "S"
2.	CPAN 1909	Tanager'S' - Siskin'S' - Pavon'S'
3.	CPAN 1955	Bb Inia, 26591, 1Y-7M-0Y-55Y-0M
4.	CPAN 1956	12300 x LV-8156/Nor-67
5.	DWL 5023	(Cr'S' -Ld"S") Gr"S"
6.	HD 2009	LR 64A x Nai 60
7.	HD 2190	HD 1954 x Son64-Tzpp-Nai 60/Cno/HD 1962-E
8.	HD 2270	4870 K.65
8.	HD 2270	HD 1962 (E 4870-Kbs/HD 2119 x 247
9.	HD 2285	249 x HD 2160/HD 2186
10.	HD 2315	HD 2160 -Tob-Ciano-23584/NA160-T.T.Son64/
11.	HD 2329	HD 1962 x E 4870 x K65/HD 1553 x UP 262
12.	HD 2428	HD 1949 x HD 2160
13.	HI 784	Napo-Tob's'/8156/Kal-Bb
14.	HI 1077	Galilo-Aust.61-151-Cno-No.66-Kal-Bb
15.	HS 207	(Kavkaz x Buhó) x (Kal x Bb)
16.	HW 741	Bb-cc/Cno-No.66/Pf 62
17.	HW 840	Tob.66 x TR 260
18.	JU-12	HDM 22550-3 x JA 3-3-1
19.	K 7410	K 812 x Kal
20.	K 8020	Kalyansona x Janak
21.	PBW 154	HD 2160-HD 2177
22.	PBW 175	HD 2160/WG 1025
23.	VL 490	NS 879/4 x Gritija
24.	VL 616	Sonalka x CPAN 1507
25.	WG 2109	Ravt 43 x HD 2177
26.	WH 147	(E 4810 x C 303) x 5339 x V 18
27.	WH 416	WH 147 x UP 368

PREVALENCE AND DISTRIBUTION OF PHYSIOLOGIC RACES OF STEM RUST
(*Puccinia graminis* *tritici*) IN PENINSULAR INDIA DURING 1986-87 TO
1988-89.

R. J. PATIL, R. T. SARKAL AND M. L. MUTKARKAR

Regional Wheat Rust Research Station, Mahabaleshwar 412 806, Dist.
Satara, Maharashtra, INDIA

SUMMARY

Analysis of stem rust samples collected from Tamil Nadu, Karnataka,
Maharashtra and Madhya Pradesh States revealed the presence of
races/biotypes 21A1, 40, 40A, 42, 42B2, 117, 117A and 117A1 in
Peninsular India during 1986 to 1989.

INTRODUCTION

Stem rust (*Puccinia graminis* f. sp. *tritici*) is considered to be the
most damaging disease of the wheat crop. Stem rust is more important
in Peninsular India and has caused severe epidemics in past. The
survey and monitoring of the rust race flora pattern of stem rust
pathogen is therefore of much importance.

MATERIALS AND METHODS

For detection of physiologic races/biotypes of wheat rust and to study
their prevalence and distribution, a survey was made using a
collection of rust affected wheat samples during the year 1986-87,
1987-88, and 1988-89 in the wheat growing areas of Tamil Nadu,
Karnataka, Maharashtra and Madhya Pradesh States. Besides these
collections, samples received from co-operators of trap nurseries were
multiplied on the susceptible wheat cultivar Pusa-4 and were analysed
on a set of Standard International Differentials, selected by Stakman
and Levine (1922) for stem rust.

RESULTS AND DISCUSSION

The prevalence and frequency pattern of different races/ biotypes of
stem rust in the different States are presented in Table 1.

Races/biotypes 40, 40A and 117 were detected from Tamil Nadu during
off season and Rabi season. Races/biotypes 21A1, 40, 40A, 42, 42B2,
117, 117A and 117A1 were prevalent in Karnataka, while races/biotypes
40A and 42B2 were prevalent in Maharashtra. In Madhya Pradesh race

21A1 was prevalent. The year 1988-89 was a rust free year for Maharashtra and Madhya Pradesh.

During 1980 to 82 Bahadur et al. (1985) reported the races/biotypes 21, 21A2, 24A, 40A, 117A, 117A1 from Peninsular India. More et al. (1985) reported races/biotypes 15C, 21, 21A1, 34A, 40A, 117, 117A, 117A1 and 122 from Peninsular India during 1981-82.

During 1984-86 Mutkekar et al (1987) reported the race flora of Peninsular India, races/biotypes 15C, 40, 40A, 42B2, 117, 117A and 122 were predominant in nature.

The present race analysis has indicated that race complex 40, 42 and 117 of stem rust occurred at the highest frequencies during 1986 to 1989 and suggest the need for deploying the genes for developing rust resistant varieties against above races.

ACKNOWLEDGEMENTS

The authors are thankful to the wheat rust Mycologist, Regional wheat Rust Research Station, Mahabaleshwar for providing facilities for the above research. Thanks are due to Dr. V.K. Shinde for critically reading the manuscript.

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Table 1. Prevalence and per cent frequencies of races and biotypes of stem rust (Puccinia graminis tritici) in Peninsular India during 1986-87 to 1988-89

State	1986-87		1987-88		1988-89	
	No. of samples analysed	Races detected	No. of samples analysed	Races detected	No of samples analysed	Races detected
1. Tamil Nadu						
(a) Off season	30	40(33.33) 40A(63.33) 117(3.33)	23	40A(100)	66	40(60.60) 40A(39.39)
(b) <u>Rabi</u> season	-	-	7	40(57.14) 40A(42.85)	-	-
2. Karnataka	30	42(3.33) 42B2(66.66) 117A(20) 117A1(6.66) 117(3.33)	6	21A1(16.66) 40(16.66) 40A(16.66) 42B2(33.33) 117A1(16.66)	13	42B2(100)
3. Maharashtra	4	42B2(100)	6	42B2(49.98) 40A(49.98)	Rust	Free year
4. Madhya Pradlesh	(Samples not received)		2	21A1(100)	Rust	Free year

Figures in parenthesis indicate per cent frequencies of prevalent races/biotypes.

FIELD AND GLASSHOUSE EVALUATION OF WHEAT TO STEM RUST

S.C. BHARADWAJ, P. BAHADUR, S.K. NAYAR AND S. NAGARAJAN

IARI Regional Station, Flowerdale, Simla-171 002, India.

Wheat is an important cereal crop and suffers every year from different diseases. Stem rust of wheat caused by *Puccinia graminis* and *Penicillaria indica* where the life of cultivars is reduced to 3-4 years due to arrival of new variants (1). A number of entries of wheat are developed by various breeders, which are evaluated for resistance to different diseases. The results of about 600 entries evaluated against stem rust of wheat in the seedling and adult are presented here.

For glasshouse evaluation, seedlings were raised in bread pens. About 7 days old seedlings were inoculated with individual races and kept in moist chamber for 48 hrs. The trays were shifted to glasshouse at a temperature of 25-30°C for incubation. Host-pathogen reaction was recorded according to Stakman et al. (4). Twenty pathotypes including - 11A (203 G 15), 14 (16 G 2), 15 (58 G 15), 17 (73 G 7), 21 (9 G 5), 21A-1 (20 G 21), 21A-2 (75 G 5), 24 (18 G 3), 24 A (5 G 19), 34 (26 G 18), 40 (104 G 13), 40 A (62 G 29), 42 (19 G 35), 42 B (7 G 35), 117 (37 G 3), 117 A (36 G 2), 117 A-1 (38 G 18), 122 (7 G 11), 184 (53 G 21) and 295 (7 G 43) were used for seedling resistance evaluation.

a) Seedling Evaluation

Entries resistant to all the pathotypes:

AKW 381, BR 2061, BR 2094, BM 11, BM 1108, BM 142, BM 146, BM 161, CPAN 1798, CPAN 1828, CPAN 1830, CPAN 1859, CPAN 1883, CPAN 1907, CPAN 1910, CPAN 1922, CPAN 1946, CPAN 1959, CPAN 1962, CPAN 1967, CPAN 1973, DL 95-2, DL 230-4, DL-253-28, DWR 59, DWR 61, HD 2009, HD 2270, HD 2278, HD 2285, HD 2297, HD 2323, HD 2329, HD 2347, HD 2367, HD 2374, HD 2379, HD 2382, HD 2402, HD 2403, HDR-2, HB 208, HB 629, HI 1968, HI 1977, HI 1048, HI 1074, HI 1121, HI 1102, HI 1452, HI 1479, HI 1487, HI 1498, HI 1518, HI 1524, HS 172, HS 207, HUH 202, HUH 206, HUH 249, HM 517, HM 741, HM 971, J 405, J 408, J 431, JNIT 67, JMJ 78-5, JMJ 76-43, JMJ 78-6, JMJ 78-8, JMJ 80-4, K 8020, K 8103, K 8110, K 8144, K 8152, K 8228, NI 8289, NI 8303, NI 8306, NI 8414, NI 8553, NI 8611, PBM 96, PBM 102, PBM 124, Raj 1555, Raj 1972, Raj 2184, Raj 2448, Raj 2525, Raj 6097, UF 262, UF 1084, UF 1110, UF 2189, VL 401, VL 421, VM 115, VM 120, VM 121, VM 147, WM 283, WL 410.

b) Adult Evaluation

The above entries were further evaluated for resistance in the field to above races of stem rust and observations recorded according to modified Cobb's Scale. Out of these CPAN 1946, DL 230-4, HD 2285,

HD 2379, HS 207 and HW 971, showed field resistance. HS 207 also provides field resistance to leaf and stripe rusts, while DL 230-4, HD 2285 and HW 971 exhibited resistance to leaf rust.

Bahadur et al. (2,3) evaluated a number of wheat entries of T. aestivum and T. durum at different hot spots. Many of the entries belong to exotic T. durum (tall wheat). The present entries include dwarf wheat. The entries providing resistance in the seedling and adult would be very useful for cultivation and breeding wheat for disease resistance.

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RESISTANCE OF THE SPRING BARLEY CV. TRIUMPH TO BARLEY BROWN RUST
(Puccinia hordei Oth.).

H.W. Roderick, E.R.L. Jones & B.C. Clifford

Institute of Grassland and Environmental Research, Welsh Plant
Breeding Station, Plas Gogerddan, Aberystwyth, Dyfed, SY23 3EB, UK.

INTRODUCTION

The spring barley cv. Triumph was grown extensively in the United
Kingdom between 1979 and 1986. Walther & Liehmann (1980) reported
that the German cv. Trumpf (syn. cv. Triumph) carried the P₉ gene for
resistance to Puccinia hordei Oth. which is found in the brown rust
differential line C.I.1243 (Udeogalanya & Clifford, 1978), but
Clifford & Jones (1981) concluded from virulence studies in the UK
that cv. Triumph carried an additional genetic factor(s).

Previous studies (Clifford & Roderick, 1978) indicated that there was
a relationship between the rate of fungal growth of avirulent and
virulent races of Puccinia hordei in barley (Hordeum vulgare L.)
genotypes in the presence of certain major (P_a) genes governing
onset of asexual reproduction with the virulent race. The rate and
extent of colony development up to this time was postulated to be a
function of the 'background' resistance and therefore probably under
different genetic control to that affecting compatibility (Parlevliet
& Kuiper, 1977). This led Clifford & Roderick (1981) to propose that
the degree of susceptibility to virulent pathogen isolates of
cultivars carrying certain major genes could be predicted, by
comparing the rate of fungal development to susceptible and partially
resistant cultivars. This technique could therefore be used to test
the theory with regard to the brown rust resistance of cv. Triumph.

This paper is an attempt to gather together information on the
resistance of cv. Triumph to brown rust and the durability of this
resistance.

MATERIALS AND METHODS

Survey of virulence frequency

Samples of brown rust infected barley leaves were sent to the Welsh
Plant Breeding Station annually between 1975 and 1991 as part of the
United Kingdom Cereal Pathogen Virulence Survey. The rust was
increased on the susceptible cv. Midas and then inoculated on to a
standard set of nine differential cultivars each possessing different
resistant gene(s) (Clifford, 1974) together with cv. Triumph and using

Virulence to the Pa9 resistance gene, present in C.I.1243, was first detected in the UK in 1975 (Table 1) as reported by Clifford & Clothier (1976) although cv. Triumph was later found to be resistant to isolates with this virulence. Although it had been reported that cv. Triumph was carrying the Pa9 gene, virulence to cv. Triumph was not detected until 1981 indicating that it carries an additional factor(s). Only one isolate out of 478 tested between 1982 and 1991 carried virulence to cv. Triumph without virulence to Pa9 (race octal 1253).

Virulence frequencies

RESULTS & DISCUSSION

colony was calculated by multiplying the length with the width. The area of each the colony and host cell responses were also noted. The area of each the longest and shortest axis were measured: the reproductive stage of colonies (single infections) were selected at random and the length of interference contrast optics. For each leaf segment, twenty rust were examined under a Zeiss RA microscope with Nomarski differential treated similarly 8 days after inoculation. The stained leaf segments using the method of Shipton & Brown (1962). The other two plants were segments were fixed and stained in alcoholic lactophenol/aniline blue from the distal and proximal parts of the inoculated leaves. The leaf were removed and two segments, approximately 25 mm in length, were cut arranged in a completely randomised design. After 6 days two plants temperature of 18±5°C, under natural daylight. The plants were pre-inoculation environment in a spore-proof glasshouse at an average chamber, incubated in the dark for 16h at 15°C and return to their isolate. The plants were then transferred to a dew stimulation settling tower. This gave a spore concentration of approximately 27 spores per mm². This procedure was repeated using a Triumph-virulent uredeospores of a Triumph avirulent isolate of P. hordei in a spore Tasman (a derivative of cv. Triumph), Vada (slow-rusting control) and Midas (susceptible control) were uniformly inoculated with 3.5 mg of The adaxial surface of the third leaf of four plants of cvs Triumph,

Experiment 2. Level of background resistance in cv. Triumph and cv. Tasman

Plants were assessed for their reaction type 15 days after inoculation. C.I.1243 (parents), and four of cv. Midas (susceptible control). Plants were assessed along with six F₁s, six each of cv. Triumph and hordei avirulent on both cv. Triumph and C.I.1243. A total of 91 F₂ Reciprocal crosses were made between the barley cv. Triumph and C.I.1243. Seeding plants were inoculated with an isolate of P.

Experiment 1. Inheritance of brown rust resistance in cv. Triumph

standard procedures of inoculation, culture and assessment (Clifford, 1977). When the symptoms were fully expressed the host response was scored using standard nomenclature.

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In summary, both pathogen virulence and genetic analysis suggest that the brown rust resistance of cv. Triumph is different from that of C.I.1243. The susceptibility of cv. Triumph to a virulent isolate was greater than would have been predicted from previous work (Cifford & Roderick, 1981). Since the brown rust resistance in the two genotypes is similar in that their expression is altered at high temperatures (above 20°C), further histological work along with a fuller genetic study should reveal much more about these resistances.

The colony areas were subjected to a square root transformation to improve the distribution of error values and thus validate the ANOVA. The mean colony sizes (Table 4) on cvs Triumph and Tasman at both 6 and 8 days after inoculation were significantly larger with the Triumph-virulent isolate than the avirulent isolate ($P < 0.001$) whereas the colonies on cvs Midas and Vada were not significantly different. Therefore, either the background resistance of cvs Triumph and Tasman has been eroded by the Triumph-virulent isolate or the major gene itself was affecting colony growth. The fact that colonies on cv. Tasman were smaller than on cv. Triumph, with both isolates, suggests that the background resistance was effective in maintaining this differential and that, in this case, the major gene was contributing to the reduced growth of colonies.

Experiment 2

Although the number of plants evaluated was rather low, recombination was observed between the two resistances. Further, when the progeny of the recombinant plants were tested there was no segregation. It therefore appears from these data that the resistances in C.I.1243 and cv. Triumph are under separate genetic control and that cv. Triumph does not carry Pa9.

There was no significant difference between the F_2 ratios of resistant and susceptible plants in reciprocal crosses ($\chi^2 = 0.687$; $0.5 > P > 0.2$) and so the data were combined for analysis. Chi-square analysis (Table 2) showed that the observed ratio differed significantly from the 15:1 ratio expected for two independent genes. However, C.I.1243 was not fully incompatible (Table 3), giving an intermediate response with some chlorotic spotting and small pustules. This variable reaction, particularly in response to temperature during the post-inoculation period, has previously been reported (Udeogalanya & Cifford, 1978). As a result plants heterozygous for Pa9 and lacking resistance from cv. Triumph could have been mis-classified as susceptible, giving a distorted ratio. Chi-square tests showed that the observed ratio most closely fitted a 13:3 which could be explained by incomplete dominance of the C.I.1243 gene (Tan, 1977).

Experiment 1

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Table 1. Percentage of isolates virulent on cv. Triumph and C.I.1243 in the UK, 1975-91

Year	Virulent on C.I.1243 (Pa9)	Virulent on cv. Triumph & C.I.1243	No. of isolates tested
1975	11	-*	9
1976	33	-	12
1977	11	-	36
1978	0	-	21
1979	0	-	26
1980	61	0	45
1981	100	4	54
1982	100	31	22
1983	100	67	21
1984	100	88	24
1985	96‡	92	26
1986	100	77	30
1987	100	88	46
1988	100	83	60
1989	100	65	73
1990	100	61	49
1991	100	93	53

* Triumph first included in tests in 1980.

‡ One isolate identified with virulence to cv. Triumph but avirulent on C.I.1243

Table 2. Chi-square analysis of the ratio of resistant and susceptible plants in an F2 population in a cross between Triumph and C.I.1243 inoculated with an isolate of *P. hordei* avirulent on the parent lines.

	R	S
Observed F ₂ ratio	77	14
Expected F ₂ ratio	85.31	5.69

$\chi^2[1]=12.94; P>0.001$

Assuming two independent dominant genes (15:1 ratio)

	R	S
Observed F ₂ ratio	77	14
Expected F ₂ ratio	73.94	17.06

$\chi^2[1]=0.676; 0.5 < P < 0.02$

Table 3. Reaction of parents and offspring to a Triumph-avirulent isolate of *P. hordei*.

Triumph	Resistant	0;
C.I.1243	Resistant	1,2-3
Midas	Susceptible	4
F1	Resistant	0;
F2	Resistant to susceptible	0/0; to 3-4

Table 4. Mean colony sizes, μm^2 , (transformed to square roots) of *P. hordei* on four barley cultivars 6 and 8 days after inoculation with either a Triumph-avrulent or -virulent isolate.

	6 days		8 days	
Triumph-avrulent isolate	0.229	0.466	0.321	0.548
Triumph-virulent isolate	0.184	0.341	0.262	0.438
Tasman	0.396	0.401	0.382	0.420
Midas	0.251	0.255	0.492	0.502
SED \pm	0.013		0.017	

The seeds of 50 accessions of *Triticum durum* were obtained from the stored world wheat germplasm at the Punjab Agricultural University Research Farm, Keylong (Himachal Pradesh). These germplasm lines were tested for seedling reaction to individual pathotypes (five to ten Indian pathotypes) of *Puccinia recondita* f.sp. *tritici*. In addition, sixteen lines carrying known leaf rust resistance genes (*Lr* isogenic lines) were also tested for seedling resistance to ten pathotypes. These lines were: Malakof (*Lr* 1), Webster (*Lr* 2a), Loros (*Lr* 2c), Democrat (*Lr* 3), *Lr* 9, *Lr* 10, *Lr* 13, *Lr* 14a, *Lr* 15, *Lr* 17, *Lr* 18, *Lr* 19, Thew (*Lr* 20), *Lr* 23, *Lr* 24 and Benno (*Lr* 26). The first leaf of 7-8 days old seedlings was inoculated following the standard

MATERIALS AND METHODS

Tetraploid species of *Triticum*, including *Triticum durum*, have been used as sources of rust resistance to provide genetic diversity in hexaploid cultivated wheat (Stakman, 1968; Watson and Luig, 1968). It has been observed in India that, in general, durums are more resistant to leaf rust as compared to hexaploid wheats. Therefore 50 accessions of *T. durum* from the germplasm collection at the Punjab Agricultural University, most of which have shown promise for leaf rust resistance in field screening tests at hot spots (Gill, 1989), were tested with individual pathotypes of leaf rust at the seedling stage. The results of this study are presented here.

INTRODUCTION

Fifty accessions of *Triticum durum* germplasm and sixteen lines carrying known leaf rust resistance genes (*Lr* gene) were tested with five to ten pathotypes of *Puccinia recondita* f.sp. *tritici*. Matching of the infection types of the test lines with that of standard *Lr* isogenic lines indicated that seven lines may possess at least one known leaf rust resistance gene (*Lr* 3, *Lr* 13 or *Lr* 23). Reaction patterns of 43 accessions could not be matched to a known *Lr* gene or a combination of *Lr* genes. Occurrence of 23 different reaction patterns among these 43 lines suggested that there is large diversity for leaf rust resistance in these germplasm lines. The usefulness of *Triticum durum* germplasm as a potential source of leaf rust resistance for breeding rust resistant *Triticum aestivum* lines is discussed.

ABSTRACT

Biotechnology Centre, Punjab Agricultural University Ludhiana-141 004, INDIA.

HARJIT SINGH, H S DHALIWAL AND K S GILL

DIVERSITY FOR LEAF RUST RESISTANCE IN *TRITICUM DURUM* GERMPLASM

Cereal Rusts and
Powdery Mildews Bulletin

Vol. 20, Parts 1 & 2 1992

procedures for inoculation of seedlings. Two weeks after inoculation, seedling reactions were recorded according to the key developed by Mains and Jackson (1926). The matching technique was used to postulate the probable presence of Lr gene or gene combinations in the T. durum lines. This technique involves the matching of the infection type of the test lines with that of the standard Lr lines for each of the pathotypes. To record field reaction to prevalent races of leaf rust, each accession was grown in 1m long double rows during the crop years 1990-91 and 1991-92.

RESULTS AND DISCUSSION

The seedling reactions of the fifty germplasm accessions of Triticum durum and selected Lr lines are given in Table 1 and Table 2. Matching of seedling reactions of the germplasm accessions with that of standard Lr lines has shown that there are four lines (Table 1 and Table 2) which may possess Lr 23 and another unknown gene(s) providing resistance to additional pathotype(s). Similarly, there are at least two other lines (Table 1) which may possess Lr 13 and another unknown gene. One accession is expected to possess Lr 3 and an unknown gene(s) for resistance to pathotypes 12-2 and 77 (Table 2). Lr 23 is reported in Gaza durum (Watson and Ludwig, 1961; McIntosh and Dyck, 1975) and Lr 13 has been implicated in some of the Indian durum varieties (Goel and Sinha, 1990). Two accessions were susceptible to all the five pathotypes used for testing (Table 1).

Among the remaining 43 accessions out of 50, there were observed 23 different patterns of seedling reactions to the individual pathotypes (Table 1 and Table 2). None of these patterns matches with that of a known leaf rust resistance gene or a combination of Lr genes. This indicates that these germplasm lines possess Lr genes that have yet to be identified/named. Zhang and Knott (1990) reported seven previously unidentified Lr genes in durum wheat cultivars. Occurrence of as many as 23 different reaction patterns among the above said 43 accessions indicates that there is large variability for leaf rust resistance genes in these lines. Therefore, these T. durum lines may be useful as a potential source of new leaf rust resistance genes to provide genetic diversity in hexaploid cultivated wheat.

Six accessions gave seedling resistance to all the pathotypes of leaf rust used in the seedling tests (Table 1 and Table 2). The field reactions of these lines at Ludhiana over two years are given in Table 3. These accessions were either free from leaf rust or had resistant pustules in traces except accession No 3176 which gave 5MR reaction in 1990-91. However, this accession was free from leaf rust in the year 1991-92. These T. durum lines could be a good source of leaf rust resistance in a bread wheat improvement programme.

The present observations indicate large intraspecific diversity for leaf rust resistance in the Triticum durum germplasm. Since, it is easier to transfer rust resistance from T. durum to T. aestivum than from a distantly related species, it may be useful to look for new unexploited genes for leaf rust resistance in the T. durum germplasm.

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This research has been financed in part by a grant made by the United States Department of Agriculture under US India Fund (Project No. IN-ARS-639; Grant No. FG-IN-739).

ACKNOWLEDGEMENTS

Table 1: Seeding reactions of germplasm accessions of Triticum durum and Lr isogenic lines to individual pathotypes of leaf rust.

Reaction to leaf rust pathotype	Reaction		Lr genes(s)	Postulated Lr genes(s)	No of accessions/Lr isogenic line
	(S.No.)/pattern	(S.No.)/Lr isogenic line			
77A-1	R	R	I	I	6
77-1	R	R	II	II	5
77-2	R	R	III	III	1
77-4	R	R	IV	IV	2
108	R	R	V	V	1
	R	R	VI	VI	1
	R	R	VII	VII	1
	R	R	VIII	VIII	2
	R	R	IX	IX	1
	R	R	X	X	1
	R	R	XI	XI	1
	R	R	XII	XII	1
	R	R	XIII	XIII	1
	R	R	XIV	XIV	2
	R	R	XV	XV	1
	R	R	XVI	XVI	3
	R	R	XVII	XVII	1
	R	R	XVIII	XVIII	2
	R	R	XIX	XIX	3
	R	R	Lr 23+@	Lr 23+@	1
	R	R	Lr 13+@	Lr 13+@	1
	R	R	Lr 13	Lr 13	1

R=Resistant reaction (0; to 2); S=susceptible reaction (3 to 4); X=Mesothetic reaction @=another gene for additional resistance to the pathotype with reaction underlined in the table.

Table 3: Field reactions of six T. durum accessions which gave resistance to all the individual pathotypes of leaf rust used for seedling tests

Accession No.	Field Reaction	1990-91	1991-92
3034	TR	F*	F
3173	F	F	F
3176	5 MR	F	F
13723	TR	F	F
13743	TR	F	F
13744	TR	F	F
Susceptible check (Agra Local)			
		80S	90S

* Free from rust

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