Inheritance of Virulence to Three Bean Cultivars in Three Isolates of the Bean Rust Pathogen

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ABSTRACT

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Two single-pustule collections of the bean rust pathogen (*Uromyces phaseoli* var. *typica*), isolates S1-5 and P10-1, gave differential reactions on two bean cultivars, US#3 and Early Gallatin (EG), while both isolates were virulent on Pinto 111. The third isolate, S1-1, was virulent on all three cultivars. Upon selfing, S1-5 did not segregate for virulence, whereas P10-1 and S1-1 segregated only on EG and US#3, respectively. Crosses were made among the three isolates in all combinations. The F_1 progeny of P10-1 × S1-5 segregated in a ratio of one virulent to one avirulent on EG and US#3, indicating two independent genes for virulence. Combining the results of selfing and of all the crosses, three genes for virulence to the three cultivars were identified. Further proof of the three virulence genes was obtained by selfing, crossing, and backcrossing selected F_1 isolates. The three virulence loci were designated as UpA_1, UpA_2, and UpV_3. For two of the genes avirulence was dominant, but for the third, virulence was dominant.

Additional key words: albino, fungal genetics, gene-for-gene, *Phaseolus vulgaris,* quantitative inoculation.

Variation in virulence in *Uromyces phaseoli* (Pers.) Wint. var. *typica* Arth., an autoecious macrocyclic rust fungus parasitic on beans (*Phaseolus vulgaris* L.), was first discovered by Harter and Zaumeyer (7). Since then, many people have reported variation in virulence (1), but the inability to induce teliospore germination precluded studies of the inheritance of virulence. Such studies are now possible through the use of Groth and Mogen's (5) technique to induce teliospore germination.

In other obligate parasites, virulence is usually inherited as a monogenic recessive trait, although some genes for virulence are dominant (2–4,8–10,13,17,19). Some virulence genes have been shown to be closely linked (2,14,18,19).

The objective of this study was to determine the mode of inheritance of virulence of three selected rust isolates on three bean cultivars.

MATERIALS AND METHODS

Three isolates of *U. phaseoli* var. *typica* were used: P10-1, a single-pustule collection from a field collection made on the cultivar Pinto 111 near Barney, ND, in August 1976, and S1-5 and S1-1, single-pustule collections from the same field collection made from the cultivar Seafarer near Saginaw, MI, in September 1976. Mass urediospore collections were increased by inoculation procedures of Groth and Shrum (6). Single-pustule isolates were established by collecting spores from isolated uredia with a small cyclone spore collector and were increased and maintained on differentially susceptible bean cultivars. Isolate P10-1 was maintained on US#3, S1-5 on Early Gallatin (EG), and S1-1 on either Pinto 111 or EG. Plants with each isolate were kept either in isolation chambers or in the greenhouse as far as possible from one another to avoid contamination.

Bean seed was germinated in vermiculite, and the seedlings were transplanted into steamed soil. They were inoculated before the unifoliolate leaves were fully expanded at half of the leaf area. Infection types of the three rust isolates on the three cultivars were rated on a scale of 0 to 9 (6). Infection types 1 and 2 were characterized by some necrosis and were designated avirulent, whereas progressively higher-numbered types 3 to 9 caused no necrosis and were designated virulent.

Groth and Mogen's technique (5) was used with minor modifications to induce teliospores to germinate and to complete the life cycle. One modification was that teliospores were concentrated on Whatman 5.5-cm-diameter glass microfiber filter papers in a Millipore filter holder, resuspended in 20 ml of distilled water, and poured on the surface of 2% water agar plates. After teliospores germinated, these plates were suspended over Pinto 111 seedlings. Isolates used as females in crosses were suspended over plants for 1–2 days to obtain isolated pycnia, whereas isolates used as donors or for selfing were suspended over plants for several days to obtain large numbers of pycnia per leaf. Plants with pycnia were kept in insect-free cages to avoid spurious fertilization.

Mass-selfing of rust pathogen isolates was done by placing a few drops of distilled water on leaves bearing abundant pycnia and rubbing the leaf surface lightly with an alcohol-rinsed, dry finger. Aecia matured 10–12 days after fertilization; the aeciospores were mass collected into No. 00 gelatin capsules and were used immediately to inoculate plants of the three cultivars. Uredial reactions were rated 14 days later, and the pustules and flecks were counted on cultivars on which segregation for reactions occurred. Two to eight mass-selfings of each isolate were used to obtain the total number of flecks and pustules segregating on a cultivar. The total numbers were evaluated in relation to likely hypothetical ratios predicted by the chi-square test for goodness of fit.

Since the genetic ratios of the mass-selfings were determined by counting pustules and flecks as estimators of aeciospores of each genotype produced, infectivity (numbers of pustules or flecks per number of fresh spores applied) was assumed to be the same for all genotypes of the rust pathogen on all cultivars. To obtain this information, uredospores of isolates P10-1 and S1-5 were used in quantitative inoculation (QI) studies on the cultivars US#3 and EG. Five replicate groups of 20 plants of each cultivar were inoculated by using a quantitative inoculator (13) that places a repeatable number of uredospores onto a circle of leaf tissue. Seventeen milligrams each of isolate P10-1 and S1-5 uredospores were placed in 17 ml of 0.3% water agar medium. While the

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uredospore suspensions were continuously mixed by a water-driven magnetic stirrer, the uredospores were deposited onto the two unifoliolate leaves of each plant. After inoculation, the plants were placed in a dark chamber for 24 hr at 100% relative humidity and then placed on a greenhouse bench. The number of pustules or flecks (type 1 or 2) of each isolate was counted on each corresponding unifoliolate leaf of each cultivar. The mean difference in number of pustules and flecks between the two isolates for each cultivar of each replicate was used to compare replicates according to Tukey's test. If no difference was indicated, the data from all replicates for each cultivar were combined. A paired *t*-test was used in the analysis of the mean differences between the two isolates from the combined data for each cultivar.

The three isolates were crossed in all six possible combinations by using another modification of Groth and Mogen's technique (5) in which sterile toothpicks were used to transfer pycniospores. Transfers were made from three donor pycnia to one isolated female pycnium. Aeciospores were collected 7–12 days later and used immediately to inoculate the three cultivars. The F_1 uredial reaction on each cultivar was recorded 14 days after inoculation. The number of times that each infection type occurred on each cultivar was compared to hypothetical ratios; then the number of classes of progeny, characterized by reactions on both differential cultivars, was recorded for each of the six crosses. The results from reciprocal crosses were combined if no large differences in the ratios of the classes of progeny were observed. The number of times that each class of progeny occurred for reciprocal crosses was compared to hypothetical ratios and evaluated with the chi-square test for goodness of fit.

One F_1 isolate was randomly selected from three of the four classes of progeny, increased, maintained on differentially susceptible cultivars, and allowed to produce telia. Each F_1 isolate was mass-selfed and evaluated as were the parental isolates. Crosses and backcrosses with the F_1 isolates were made by using the same techniques as those used for the parents.

RESULTS AND DISCUSSION

Isolate P10-1 was virulent (infection type 7) on both Pinto 111 and US#3, but was avirulent (infection type 1) on EG. Isolate S1-5 was virulent on Pinto 111 and EG with infection types 7 and 4, respectively, but was avirulent on US#3 with an infection type 2. Isolate S1-1 was virulent on all three cultivars with an infection type 7 and Pinto 111 and US#3 and 4 on EG.

The S_1 (first self) on P10-1 were homozygous virulent on Pinto 111 and US#3, but segregated on EG with 4,944 flecks:1,576 pustules, fitting expected ratios for a single gene segregating ($\chi^2 = 3.59$, $P = 0.05–0.1$) with avirulence dominant. The S_1 of S1-5 were homozygous virulent on Pinto 111 and EG, but homozygous avirulent on US#3. The S_1 of S1-1 were homozygous virulent on Pinto 111 and EG, but segregated on US#3 with 547 flecks:1,543 pustules fitting expected ratios for a single gene segregating ($\chi^2 = 1.5$, $P = 0.10–0.25$) with virulence dominant.

The S_1 of P10-1 also segregated for color of uredia giving a mixture of both the normal brown uredia and albino uredia. The percentage of albino uredial color varied from about 10 to 25%, suggesting most likely one gene segregating for color with the albinism due to a recessive allele. The variability of percentage of albino seemed to depend on environmental conditions and would most likely be due to reduced fitness compared to the brown type.

QI studies with P10-1 and S1-5 were replicated five times on EG giving an average of 547 flecks and 540 pustules, respectively. The difference was not statistically significant ($P<0.50$). The results of the five replicates for US#3 were combined for the paired *t*-test even though one replicate was significantly different from the other four. The reason they were combined was that there was no difference in the *P*-value of the paired *t*-test with or without the significantly different replicate. In five replications on US#3 there was an average of 677 pustules of P10-1 and 867 flecks of S1-5. The difference was not statistically significant ($P<0.50$). The results of the QI studies indicate that infectivity (number of uredial pustules or flecks per number of uredospores applied) of P10-1 and S1-5 was not significantly different on either US#3 or EG. Thus, the counts of S_1 pustules and flecks should represent an unbiased measure of the number of virulent and avirulent aeciospores applied to the leaf. This method should be checked by using aeciospores rather than uredospores, but we were unable to obtain sufficient numbers of fresh aeciospores to complete such studies.

The mass-selfing technique is especially useful for studying the bean rust pathogen since the ease of obtaining pycnia and the large surface of unifoliolate bean leaves allow large numbers of pycnia to be randomly fertilized. This method differs slightly from the methods used by others (11) in that larger numbers (minimum 10^3) of pycnia are involved in the intermixing of pycniospores, and the aeciospores are mass collected and mass-transferred during inoculations. From the results of the QI studies, we assume that a mass collection of and inoculation with aeciospores should give a reasonably accurate representation of the ratios of the virulence genotypes applied to the leaf surface.

Progeny of reciprocal crosses of P10-1 × S1-5 segregated into ratio of 1:1:1:1 doubly avirulent:virulent only on EG:virulent only on US#3:doubly virulent (Table 1). The data indicate that two loci are segregating, one locus governing virulence to US#3 and the other governing virulence to EG designated as $Up A_1$ and $Up A_2$, respectively (abbreviated A_1,A_2) (16). The ratio of one avirulent: one virulent on each cultivar indicates that there is one isolate homozygous and the other heterozygous at both the A_1 and A_2 loci. Comparing the results from the crosses with the results of selfing indicates that P10-1 is the isolate homozygous virulent at the A_1 locus and heterozygous with avirulence dominant at the A_2 locus. Therefore, isolate S1-5 is homozygous virulent at the A_2 locus and heterozygous at the A_1 locus. The S_1 progeny of isolate S1-5 were homozygous virulent at the A_2 locus, which agrees with the results of the crosses, but the S_1 progeny were homozygous avirulent at the A_1 locus, which contradicts the results of the crosses. Since avirulence masks other gene-for-gene interactions due to epistasis (12), the simplest genetic interpretation is that there is a second locus governing virulence to US#3, which can be designated as $Up V_3$ (abbreviated V_3). Under this assumption, S1-5 is homozygous recessive for avirulence at the V_3 locus but heterozygous at the A_1 locus. Isolate P10-1 accordingly is homozygous dominant for virulence at the V_3 locus since no segregation was seen in the S_1 progeny. With these conclusions, the proposed genotypes of P10-1 and S1-5 are $a_1a_1A_2a_2V_3V_3$ and $A_1a_1a_2a_2v_3v_3$, respectively.

Progeny from the reciprocal crosses of S1-5 × S1-1 segregated in a ratio of 1:1 for virulent only on EG:doubly virulent (Table 1). The data indicate that segregation occurred only at the A_1 locus with isolate S1-5 being heterozygous and S1-1 being homozygous for virulence. The segregation that was observed in the S_1 progeny of S1-1 cannot be explained by either the A_1 or V_3 locus since virulence should be dominant in S1-5 if segregation in S1-1 was due to the A_1 locus, and a ratio of three avirulent to one virulent should be obtained in the F_1 progeny upon crossing S1-5 × S1-1 if segregation was due to the V_3 locus. This indicates that S1-1 probably contains another locus whose alleles affect virulence to US#3.

Progeny from the reciprocal crosses of P10-1 × S1-1 segregated

TABLE 1. Segregation ratios of F_1 progeny from crosses among those bean rust pathogen isolates expressing virulence on bean cultivars US#3 and Early Gallatin

Reaction on bean cultivar		Crosses of rust pathogen isolates					
		P10-1 × S1-5		S1-5 × S1-1		P10-1 × S1-1	
US#3	Early Gallatin	Observed	Expected	Observed	Expected	Observed	Expected
A[a]	A	50	41.5			22	21.5
A	V	36	41.5	54	52	22	21.5
V	A	35	41.5			68	64.5
V	V	45	41.5	50	52	60	64.5
Theoretical ratio		1:1:1:1		1:1		1:1:3:3	
P-value[b]		0.25–0.50		0.50–0.75		0.90–0.95	

[a] A = avirulent; V = virulent

[b] Based on the chi-square test for goodness of fit.

in a ratio of 1:1:3:3 doubly avirulent:virulent only on EG:virulent only on US#3:doubly virulent (Table 1). The data indicate that there are two loci. The ratio of one avirulent to one virulent on EG was expected, since previous results show P10-1 to be heterozygous and S1-1 to be homozygous virulent at the A_2 locus. The ratio of one avirulent to three virulent on US#3 was not expected since it would indicate that both isolates are heterozygous at one locus governing virulence to US#3, whereas P10-1 is known to be homozygous virulent on US#3. Data from this cross gave further evidence of P10-1 being heterozygous at the A_2 locus and also gave further evidence for another locus in S1-1 affecting virulence to US#3.

From these results, the proposed genotypes of P10-1, S1-5, and S1-1 are $a_1a_1A_2a_2V_3V_3$, $A_1a_1a_2a_2v_3v_3$, and $a_1a_1a_2a_2V_3V_3$, respectively, where avirulence is dominant at both the A_1 and A_2 loci, but recessive at the V_3 locus. Recent studies (2–4,8–10,12,15,18) of the genetics of virulence of obligate parasites have also shown virulence to be inherited as a dominant trait. Since S1-1 may have other loci affecting virulence to US#3, the genotype of S1-1 is more tentative.

The four types of virulence patterns observed in the F_1 progeny of the cross P10-1 × S1-5 and the reciprocal cross would result from the proposed genotypes of P10-1 and S1-5 if the A_1 and A_2 loci are independent. The V_3 locus will be considered later. Therefore, the expected genotypes of the four types of progeny are: $A_1a_1a_2a_2$, $a_1a_1A_2a_2$, $A_1a_1A_2a_2$, and $a_1a_1a_2a_2$.

To provide further evidence about the genotypes of the parental isolates, several selected F_1 progeny from the reciprocal crosses of P10-1 × S1-5 representing three of the four phenotypic classes of progeny were selfed and crossed or backcrossed. The F_1 isolates (and their genotypes) selected were designated 151 ($a_1a_1A_2a_2$), 20 ($A_1a_1a_2a_2$), and 108 ($A_1a_1A_2a_2$). F_1 isolate 151, being phenotypically a parental type similar to P10-1, was selected from a cross in which S1-5 was the female. Isolate 20, being phenotypically a parental type similar to S1-5, was selected from a cross in which P10-1 was the female. Isolate 108, being a nonparental phenotype, avirulent on both US#3 and EG, was also selected from a cross where P10-1 was the female. A representative of the fourth phenotypic class of progeny, virulent on both US#3 and EG, was not selected since it was not possible to determine if the progeny was a result of a successful cross according to the original hypothesis including only the A_1 and A_2 loci. If the V_3 locus is included in the hypothesis, an isolate could be selected from a cross in which S1-5 was the female. Each F_1 isolate was selected as previously stated to insure that it was obtained from a successful cross and not a self.

The S_1 of 151 did not segregate on 111, but did segregate on US#3 and EG with 1,440 flecks:4,715 pustules, and 3,839 flecks:1,403 pustules, respectively. While fits to the expected ratios of one locus segregating were not close ($\chi^2 = 8.88$, $P<0.005$ and $\chi^2 = 8.71$, $P<0.005$, respectively), neither did the data more closely fit any other hypothetical ratio. The segregation for one virulence locus on EG was expected according to the original hypothesis. The segregation on US#3 was expected only if the V_3 locus, where virulence is dominant, was segregating. The segregation on US#3 gave further evidence of the existence of the V_3 locus and would suggest that all isolates of the F_1 progeny should be heterozygous at the V_3 locus.

The S_1 progeny of isolate 20 segregated only on US#3 (5,412 flecks:2,232 pustules) with $P<0.005$ for a 3:1 ratio. The S_1 of isolate 108 segregated on both US#3 and EG (3,293 flecks:1,271 pustules, and 4,218 flecks:1,243 pustules, respectively) with $P<0.005$ for a 3:1 ratio. While the data did not give good fits to the expected ratio of one locus segregating, neither did the data more closely fit any other readily hypothesized ratio. The segregation observed for these two isolates was expected according to the original hypothesis and the isolates' phenotypes.

The results of the mass-selfings of the F_1 isolates were consistent with the proposed genotypes of the parental isolates and the resulting F_1 isolates' phenotypes. Even though the data gave poor fits to expected ratios, there was evidence for at least one locus segregating on the cultivars where segregation occurred. When comparing the observed number of pustules and flecks to expected ratios, there was a tendency for pustules to be overrepresented in the observed progeny. The discrepancy could be due to contamination from other rust isolates in the greenhouse; to factors other than virulence, which may have resulted in nonrandom mating of the segregating haploid genotypes; to differences in infectivity of pustules and flecks under crowded conditions on leaf surfaces, resulting in selection for virulent types; to directional errors in counting (the pustules are much more visible); or to differential effects of environmental conditions on flecks versus pustules, such as the alteration of the avirulent phenotype by high temperatures, which occurred in late spring when the results were taken for the F_1 isolates. It was observed that moderately high temperature (30–35 C) increased the pustule size and decreased the area of necrosis of infection type 2, and for infection type 1 the necrosis was sometimes barely visible.

The cross of F_1 isolates 151 × 20 was made to provide further evidence whether the A_1 locus is heterozygous in isolate 20 and homozygous in isolate 151, and whether the A_2 locus is heterozygous in isolate 151, but homozygous in isolate 20. Backcross S1-5 × 151 was made to provide further evidence whether S1-5 is heterozygous at the A_1 locus.

Progeny from cross 151 × 20 and from backcross S1-5 × 151 each segregated in a ratio of 1:1:1:1 doubly avirulent:virulent only on EG:virulent only on US#3:doubly virulent (Table 2). Along with the data of selfing the F_1 isolates, these data indicate that isolate 151 is homozygous virulent at the A_1 locus, but heterozygous with avirulence dominant at the A_2 locus; whereas isolate 20 is heterozygous with avirulence dominant at the A_1 locus, but homozygous virulent at the A_2 locus; and S1-5 is heterozygous with avirulence dominant at the A_1 locus. Not enough progeny were observed from the backcrosses to make any conclusions but in conjunction with the other cross the results help to support the original hypothesis.

The cross of F_1 isolates 151 × 108 was made to provide further evidence as to whether both the A_1 and A_2 loci are heterozygous in isolate 108. The progeny from this cross segregated in a ratio of 3:1:3:1 for doubly avirulent:virulent only on EG:virulent only on US#3:doubly virulent (Table 2). Along with the data of selfing and the cross of 151 × 20, these data indicate that isolate 108 is heterozygous at both the A_1 and A_2 loci with avirulence dominant.

The backcross P10-1 × 108 was made to provide further evidence whether P10-1 is heterozygous at the A_2 locus. The progeny of this cross segregated in a ratio of 1:1:3:3 for doubly avirulent:virulent only on EG:virulent only on US#3:doubly virulent (Table 2). These

TABLE 2. Segregation ratios of progeny from crosses and backcrosses of F_1 bean rust isolates expressing virulence on bean cultivars US#3 and Early Gallatin

Reaction on bean cultivars		Crosses of rust isolates							
		151 × 20		S1-5 × 151		151 × 108		P10-1 × 108	
US#3	Early Gallatin	Observed	Expected	Observed	Expected	Observed	Expected	Observed	Expected
A[a]	A	29	31	5	6	41	46.5	5	4.8
A	V	35	31	5	6	21	15.5	5	4.8
V	A	22	31	5	6	41	46.5	15	14.6
V	V	38	31	9	6	21	15.5	14	14.6
Theoretical ratio		1:1:1:1		1:1:1:1		3:1:3:1		1:1:3:3	
P-value[b]		0.10–0.25		0.50–0.75		0.10–0.25		0.75–0.90	

[a] A = avirulent; V = virulent.
[b] Based on the chi-square test for goodness of fit.

TABLE 3. Comparison of expected to observed ratios of segregation of bean rust pathogen F_1 progeny if the $Up A_1$ and $Up V_3$ loci in the isolates are linked or independent

F_1 Progeny of	Ratios on US#3 if:		Ratio that best fits observed
	No linkage	Linked	
P10-1 × S1-5	1A:1V[a]	1A:1V	1A:1V
151 × 20	5A:3V	1A:1V	1A:1V
151 × 108	5A:3V	1A:1V	1A:1V
S1-5 × 151	1A:1V	1A:1V	1A:1V
P10-1 × 108	1A:1V	1A:1V	1A:3V

[a]A = avirulent; V = virulent.

TABLE 4. Comparison of expected results if the $Up A_1$ and $Up V_3$ loci in the bean rust pathogen isolates are linked or independent of the observed patterns of segregation of the bean rust S_1 progeny

S_1 Progeny of	Ratios of US#3 if:		Ratio that best fits observed
	No linkage	Linked	
S1-5	HA[a]	HA	HA
P10-1	HV	HV	HV
151	3V:1A[b]	3V:1A	3V:1A
20	13A:3V	3A:1V	3A:1V
108	13A:3V	3A:1V	3A:1V

[a]HA = homozygous avirulent; HV = homozygous virulent.
[b]V = virulent; A = avirulent.

data indicate that both isolates are heterozygous with virulence dominant at the A_1 locus and only one isolate was heterozygous at the A_2 locus. This is not what was expected based on the simplest genetic hypothesis according to which P10-1 was thought to be homozygous virulent at the A_1 locus and both isolates heterozygous with avirulence dominant at the A_2 locus. This contradiction may be due to our having tested barely enough progeny to distinguish between a 1:1 or 3:1 ratio (11).

In all of the crosses the progeny segregated as expected with the A_1 and A_2 loci being independent, but this did not account for the V_3 locus. The existence of the V_3 locus was shown by the segregation of the S_1 progeny of isolate 151. The other F_1 isolates should also be heterozygous at the V_3 locus, but the results indicated only one locus segregating instead of two. The two types of progeny expected from the cross of P10-1 × S1-5 or the reciprocal cross on US#3 would result when the A_1v_3 alleles being inherited from S1-5 combined with the a_1V_3 from P10-1, and when the a_1v_3 alleles inherited from S1-5 combined with the a_1V_3 alleles from P10-1. If the A_1 and V_3 loci are totally linked with virulence alleles in coupling as in isolate 20 or 108 in which A_1v_3 alleles are inherited from S1-5 and a_1V_3 alleles are from P10-1, the results of selfing would only show the one locus segregating on US#3. The ratios expected with the A_1 and V_3 loci being independent differ from those expected when A_1 and V_3 are linked, but the difference between the two ratios is small, and is impossible to distinguish with the numbers of progenies obtained from crossing (Table 3) and from mass-selfing (Table 4). At this time it is impossible to tell whether A_1 and V_3 are linked or independent, but the data slightly more closely support the possibility that A_1 and V_3 may be linked.

Linked virulence genes are thought to be infrequent, but this may be due to the paucity of genetic studies with parasites. The results of several studies (3,14,18,19), including some involving three different obligate parasites, have indicated linked virulence genes.

The results from all the crosses except the backcross of P10-1 ×108 gave evidence that supported the original hypothesized genotypes of isolates P10-1 and S1-5. The results of all the mass-selfings also support the hypothesized genotypes. Therefore, the proposed genotypes of P10-1, S1-5, S1-1, 151, 20, and 108 are $a_1a_1A_2a_2V_3V_3$, $A_1a_1a_2a_2v_3v_3$, $a_1a_1a_2a_2V_3V_3$, $a_1a_1A_2a_2V_3v_3$, $A_1a_1a_2a_2V_3v_3$, and $A_1a_1A_2a_2V_3v_3$, respectively.

In conclusion, three virulence genes $Up A_1$, $Up A_2$, and $Up V_3$ were found in the three rust pathogen isolates. For two of them avirulence was dominant, but for the third virulence was dominant.

LITERATURE CITED

1. Ballantyne, B. 1974. Resistance to rust (*Uromyces appendiculatus*) in beans (*Phaseolus vulgaris*). Proc. Linn. Soc. NSW 98:107-121.
2. Green, G. J. 1964. A color mutation, its inheritance, and the inheritance of pathogenicity in *Puccinia graminis* Pers. Can. J. Bot. 42:1653-1664.
3. Green, G. J. 1965. Inheritance of virulence in oat stem rust on the varieties Sevnothree, Richland, and White Russian. Can. J. Genet. Cytol. 7:641-650.
4. Green, G. J. 1966. Selfing studies with races 10 and 11 of wheat stem rust. Can. J. Bot. 44:1255-1260.
5. Groth, J. V., and Mogen, B. D. 1978. Completing the life cycle of *Uromyces phaseoli* var. *typica* on bean plants. Phytopathology 68:1674-1677.
6. Groth, J. V., and Shrum, R. D. 1977. Virulence in Minnesota and Wisconsin bean rust collections. Plant Dis. Rep. 61:756-760.
7. Harter, L. L., and Zaumeyer, W. J. 1941. Differentiation of physiologic races of *Uromyces phaseoli typica* on bean. J. Agric. Res. 62:717-731.
8. Johnson, T. 1954. Selfing studies with physiologic races of wheat stem rust, *Puccinia graminis* var. *tritici*. Can. J. Bot. 32:506-522.
9. Kao, K. N., and Knott, D. R. 1969. The inheritance of pathogenicity to races 111 and 29 of wheat stem rust. Can. J. Genet. Cytol. 11:266-274.
10. Luig, N. N., and Watson, I. A. 1961. A study of inheritance of pathogenicity in *Puccinia graminis* var. *tritici*. Proc. Linn. Soc. NSW 86:217-229.
11. Miah, M. A. J. 1968. Method of selfing F_1 cultures and interpreting genetic data in rust fungi. Can. J. Genet. Cytol. 10:613-619.
12. Person, C., and Mayo, B. M. E. 1974. Genetic limitations on models of specific interactions between a host and its parasite. Can. J. Bot. 52:1339-1347.
13. Samborski, D. J., and Dyck, P. L. 1968. Inheritance of virulence in wheat leaf rust on the standard differential wheat varieties. Can. J. Genet. Cytol. 10:24-32.
14. Samborski, D. J., and Dyck, P. L. 1976. Inheritance of virulence in *Puccinia recondita* on six backcross lines of wheat with single genes for resistance to leaf rust. Can. J. Bot. 54:1666-1671.
15. Schein, R. D. 1964. Design, performance and use of a quantitative inoculator. Phytopathology 54:509-513.
16. Sidhu, G., and Person, C. 1971. Genetic control of virulence in *Ustilago hordei*. II. Segregations for higher levels of virulence. Can. J. Genet. Cytol. 13:173-178.
17. Statler, G. D. 1977. Inheritance of virulence of culture 73-47 *Puccinia recondita*. Phytopathology 67:906-908.
18. Statler, G. D. 1979. Inheritance of virulence of *Melampsora lini* Race 218. Phytopathology 69:257-259.
19. Statler, G. D. 1979. Inheritance of pathogenicity of culture 70-1, Race 1, of *Puccinia recondita tritici*. Phytopathology 69:661-663.