Gene-for-Gene Interactions During Primary Infection of Wheat by Erysiphe graminis f. sp. tritici

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ABSTRACT

Two cultures of *Erysiphe graminis* f. sp. *tritici* (MS-1 and MS-76) which possess the corresponding *P1* and *p1* genes, respectively, were used to study the development of elongating secondary hyphae (ESH) on wheat lines containing either *Pm1* or *pm1*. Development of ESH with the *p1/Pm1* genotype, a compatible combination, was similar to that observed with compatible combinations in the absence of specific *Pm* genes (*p1/pm1* or *P1/pm1*). The formation of ESH was inhibited only with the *P1/Pm1* genotype. Phytopathology 60:1068-1070.

Additional key words: Wheat, *Triticum aestivum*, powdery mildew, host-parasite interactions, genetics of primary infection.

Flor (4, 5) first formulated the gene-for-gene hypothesis as the genetic basis of host-parasite compatibility for the flax rust disease. Since this initial report, similar gene-for-gene relationships have been found with many different diseases involving both obligate and nonobligate parasites (11). The bases for many of the demonstrations of the existence of a gene-for-gene relationship rest on the relative infection types produced following inoculation of different particular host lines with various strains of the pathogen. This comparative method has been satisfactory for differentiating gross qualitative differences with various incompatible parasite/host genotypes, but the measurement of final infection type is not very useful for determining when incompatibility is specified in the ontogeny of disease.

A quantitative system for studying powdery mildew development during the primary stages of infection of wheat and barley has been reported (8, 10). These studies demonstrated that, with the appropriate environmental conditions, mildew development proceeds in a very predictable manner. A population of spores applied to the leaf surface undergo morphogenetic changes during the infection process with a high degree of synchrony. The production of elongating secondary hyphae (ESH) by the fungus was used to indicate the establishment of a successful host-parasite relationship, since the formation of ESH is dependent on the formation of functional haustoria in the host epidermal cells (9). The inhibition of mildew development with incompatible parasite/host genotypes could be quantitatively studied during primary infection by determining the percentage of applied conidia that formed ESH (9, 13, 14).

The purpose of this study was to determine whether the production of ESH corresponded to the predictions of the gene-for-gene hypothesis as applied in the "quadratic check" proposed by Rowell et al. (12). We were specifically interested in determining whether the three different compatible genotypes, involving a single gene for reaction in the host and the corresponding gene for pathogenicity in the pathogen (*P1/r1, p1/r1, p1/R1*), were similar during primary infection.

MATERIALS AND METHODS.—*Genetic symbols.*—The combinations of wheat lines and strains of the mildew fungus in this study are referred to by the known gene differences involved in each parasite/host combination. The symbols *P* and *p* (7) are used to designate the alternate alleles in the pathogen; the symbols *Pm* and *pm* (2) are used to designate the alternate alleles in the host. The combined parasite/host genotype, the genotype of the "aegricorpus" (7), specifies the particular corresponding genes which determine the progress of disease development. *Px/Pmx* parasite/host genotypes are incompatible combinations; *Px/pmx*, *px/pmx*, and *px/Pmx* parasite/host genotypes are compatible combinations. The host genotype is abbreviated in this study as *Pmx* rather than as a diploid *Pmx Pmx*, since the wheat lines used are homozygous for these genes.

Near-isogenic wheat lines.—Four near-isogenic wheat lines each containing a *Pm1* were obtained from L. W. Briggle. The genetic data and suggested *Pm* terminology for these lines have been published (1, 2, 3). The parental source of each *Pm1* gene follows the *Pm1* designation in this study, since the distinctness of the genes derived from the various parents is uncertain. Near-isogenic wheat lines possessing a *Pm1* gene were derived following eight backcrosses of the particular *Pm* gene involved to the cultivar Chancellor (Cc), *Triticum aestivum* L. 'Chancellor'. Chancellor wheat (C.I. 12333) (Cereal accession number of CRD, ARS, USDA) contains no known genes restricting mildew development and is designated as *pm1*(Cc).

Mildew cultures.—Cultures MS-1 and MS-76 of *Erysiphe graminis* f. sp. *tritici* were collected in Michigan and purified by three successive single pustule isolations from lightly inoculated Little Club wheat. Based on the resulting infection types following inoculation of the *Pm1* wheat lines, culture MS-1 possessed

the corresponding *P1* gene for incompatibility with *Pm1*; culture MS-76 possessed the corresponding *p1* gene for compatibility with *Pm1*.

Environmental conditions necessary for the production of uniform conidia have been published (10). Mildew cultures were periodically checked for uniformity by observation of fungus development during primary infection and by the resulting infection type following inoculation of a set of 13 near-isogenic wheat lines also obtained from L. W. Briggle.

Inoculation procedures and observations of fungus development.—Single wheat leaves were inoculated by the "rolling method" (10). A cotton swab was used to obtain a uniform distribution of 100-300 single conidia/cm length on the abaxial leaf surface. Fungus development was examined at various times after inoculation with a light microscope (×160). Morphogenesis of the fungus was observed on a 1- to 2-cm leaf section, and the percentage of the parasite population in each stage of development was recorded. After examination, each plant section was discarded and a new section from another plant was used for subsequent observations. Approximately 100-125 parasitic units were counted on each leaf section at each time of observation. Experiments were repeated on at least 4 different days to minimize variations in the plants or the inoculum.

RESULTS.—*General observations.*—Development of *E. graminis tritici* during the stages of germination, formation of appressorial initials, and formation of "mature" appressoria were similar with both compatible (*P1/pm1*, *p1/pm1*, and *p1/Pm1*) and incompatible (*P./Pm1*) parasite/host genotypes. The similarity in the initial stages of infection with *Pm1* or *pm1* combinations is consistent with other reports that incompatibility to powdery mildew development is expressed only after the formation of appressoria (6, 14, 15). The infection type at 7 days after inoculation with *P1/Pm1* incompatible genotypes was of type 1 (fleck). The three compatible genotypes (*P1/pm1*, *p1/pm1*, and *p1/Pm1*) were characterized by type 4 (fully compatible) infection types, and were morphologically indistinguishable.

Incompatible P1/Pm1 genotypes.—Production of ESH with the incompatible *P1/Pm1* parasite/host genotypes was shown in an earlier study (14) to be strongly inhibited. The incompatible genotypes involving *Pm1* are discussed here to complete the genetic scheme (12) and are shown in Fig. 1. The production of ESH with the *P1/Pm1* genotypes is 60-70% lower than with the compatible *P1/pm1*(Cc) genotype. This greatly inhibited formation of ESH at 26 hr after inoculation is consistent with the resulting low infection type at 7 days after inoculation.

Compatible parasite/host genotypes.—The production of ESH with the compatible parasite/host combination MS-1 and Chancellor (*P1/pm1*[Cc]) was essentially identical to ESH development with the compatible MS-1 and Little Club (*P1/pm1*[L. Club]) parasite/host combination (Fig. 2-A). This result indicated that the optimum environmental conditions determined using MS-1 and Little Club (8, 10) were satisfactory with other compatible parasite/host genotypes.

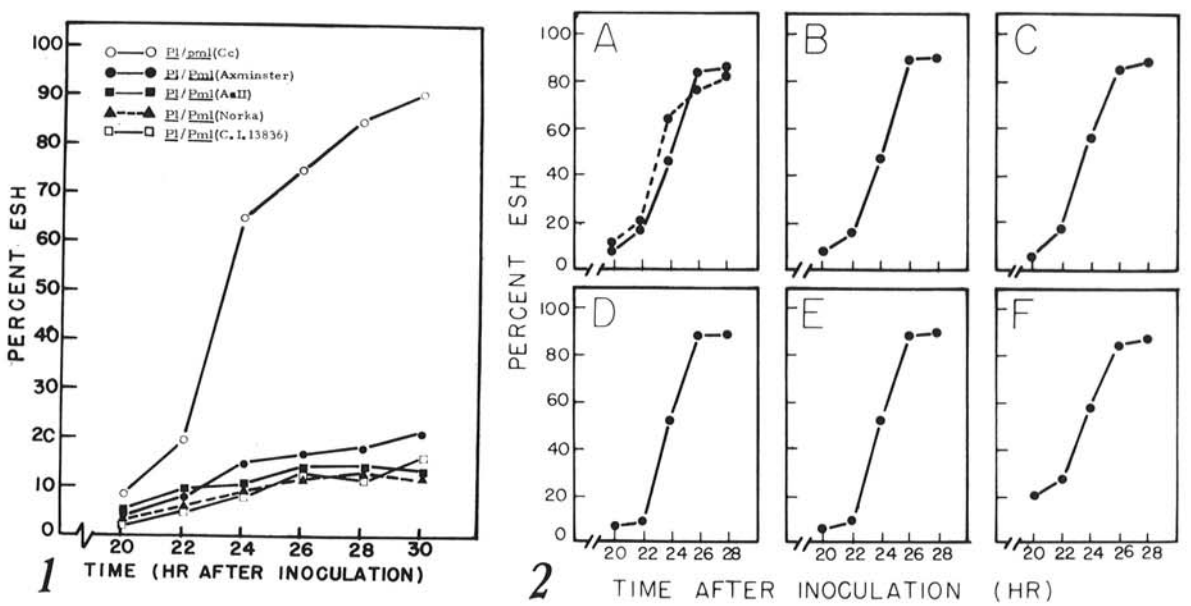

Fig. 1-2. **1)** Formation of elongating secondary hyphae (ESH) by *Erysiphe graminis* f. sp. *tritici* (MS-1) with the following incompatible parasite/host genotypes: *P1/pm1*(Chancellor) (compatible control) (○——○), *P1/Pm1*(Axminster) (●——●), *P1/Pm1*(AsII) (■——■), *P1/Pm1*(Norka) (△- - -△), and *P1/Pm1*(C.I. 13836) (□——□). **2)** Similarity of production of elongating secondary hyphae (ESH) with the following compatible parasite/host genotypes involving *Pm1*. (A) *P1*(MS-1)/*pm1*(Little Club) (●——●) and *P1*(MS-1)/*pm1*(Chancellor) (●- - -●), (B) *p1/pm1*(Chancellor), (C) *p1/Pm1*(Axminster), (D) *p1/Pm1*(AsII), (E) *p1/Pm1*(Norka), and (F) *p1/Pm1*(C.I. 13836).

ESH production with the combination of isolate MS-76 and Chancellor (*p1/pm1*[Cc]) (Fig. 2-B) was indistinguishable from that with the *P1/pm1*(Cc) genotype (isolate MS-1 and Chancellor) (Fig. 2-A). The similarity of ESH production with these two genotypes indicated that, although another culture of the pathogen was used, mildew development was not inhibited if the genotype of the aegricorpus was for compatibility. The parasite/host genotypes involving the *pml* allele were essentially identical with respect to pathogen development during primary infection and to resulting infection type.

With the *p1/Pm1* compatible genotypes (Fig. 2-C, D,E,F), production of ESH was indistinguishable from ESH production with the *P1/pm1* or *p1/pm1* compatible genotypes. Neither a quantitative nor qualitative inhibition of fungus development was observed when the *Pm1* gene for incompatibility in the wheat plant was matched with the corresponding *p1* gene in the fungus. The gene-for-gene interactions specifying compatibility in the 3 genotypes studied allowed the same development of the powdery mildew fungus.

DISCUSSION.—The genetic control of ESH production during primary infection apparently parallels the genetic control of final infection type. This correspondence to the predictions of the gene-for-gene relationship indicates that production of ESH serves as a valid criterion of parasite/host compatibility. Similarity of the three compatible genotypes studied quantitatively demonstrates that the *Pm1* gene in the host inhibits mildew development only when matched with the corresponding *P1* gene in the pathogen. The specific biochemical systems which operate to inhibit disease development with the *P1/Pm1* genotype must be either absent or inactive with *p1/Pm1* genotype. Preliminary evidence obtained by observing ^{35}S transfer from wheat to the external fungus mycelium suggests that the *p1/Pm1* compatible genotype is in fact distinct from the compatible *P1/pm1* and *p1/pm1* genotypes (R. S. Slesinski, *unpublished data*). This result suggests that the *p1* allele in the pathogen represents an altered specificity from *P1* which no longer interacts with the systems specified by the *Pm* gene to the extent of morphologically inhibiting disease development.

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