Development of Puccinia graminis f. sp. tritici on Resistant and Susceptible Barley Cultivars

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ABSTRACT

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Infection types 3-4 developed on the barley cultivar, Hiproly, inoculated with each of three races of *Puccinia graminis* f. sp. *tritici,* and infection type 1 developed on cultivars Larker and Manker. Numbers of uredia per square centimeter of leaf surface were fewer in Larker and Manker than in Hiproly. There were no significant differences between resistant and susceptible cultivars in urediospore germination, appressorium formation, or penetration. Growth of the pathogen was restricted inside the leaves of resistant barley cultivars, but not in those that were susceptible.

The "T" gene has been widely used, since the mid 1930's, to develop barley cultivars resistant to stem rust (5, 6, 7). Today it appears to be as effective as when first introduced. To ascertain the nature of this resistance, observations were made on the development of *Puccinia graminis* Pers. f. sp. *tritici* Eriks. and E. Henn. on resistant and susceptible barley cultivars (*Hordeum vulgare* L.).

MATERIALS AND METHODS

The cultivars used were: Larker and Manker, both of which have the "T" gene, and Hiproly, which does not. The plants were grown in a 21 C greenhouse. When the first leaf was fully developed, about 8 days after planting, the plants were inoculated with urediospores.

Races Hjc, TLM, and HDL, of *P. graminis* f. sp. *tritici,* obtained from the Cereal Rust Laboratory, St. Paul, Minnesota, were used. They were maintained and urediospores were increased on the Hiproly cultivar.

Plants were inoculated by rubbing them with a flat inoculating needle that had been dipped in freshly collected urediospores. After inoculation, the plants were placed in a moist chamber and kept continuously wet at approximately 24 C. The plants were kept in darkness for the first 12 hours, then in about 4,304 lux (400 ft-c) of light from fluorescent lamps for 12 hours. After 24 hours, the chamber was partially opened to permit slow drying of the plant surfaces. The plants were then transferred to a 24-27 C greenhouse.

Development of the stem rust fungus was observed 4, 8, 12, and 24 hours after inoculation. Leaf segments 2.5 cm long were sprayed with a staining solution made up from 0.3 ml of 2% acid fuchsin, 0.3 ml of 2% cotton blue, 4 ml of 1.25% acetic acid, and 18 ml of 95% ethyl alcohol (1). Germination of urediospores, formation of appressoria, and penetration (as judged by the presence of empty appressoria) were evaluated on each leaf segment.

Growth of the rust fungus in leaf tissue was evaluated 96 hours after inoculation. For this, leaf segments cut from inoculated leaves were cleared and stained according to the methods of McBryde (4) and Ashagari (2) and mounted in Permount on glass slides. Fungal colony length and width was measured with an ocular micrometer.

Data were analyzed statistically using analysis of variance.

RESULTS

Infection type and number of uredia per square centimeter.—In two trials, 11 days after inoculation, infection types were evaluated on 20 inoculated plants of each cultivar according to the system of Stakman et al. (10). Infection types 3-4 developed on Hiproly with each race, and infection type 1 developed on Larker and Manker infected with each race except for a few of type 2 on Manker infected with race TLM (Table 1).

TABLE 1. Infection type and number of uredia of races of *Puccinia graminis* f. sp. *tritici* on three barley cultivars

Cultivar	Rust race	Infection type	Uredia/cm^2[a] (mean no.)
Hiproly	Hjc	3-4	8.9 y
	TLM	3	9.0 y
	HDL	3	8.9 y
Larker	Hjc	1-c	1.7 x
	TLM	1	1.5 x
	HDL	1-c	1.9 x
Manker	Hjc	1-c	1.2 x
	TLM	1-2	1.2 x
	HDL	1	1.9 x

[a]Means followed by different letters are significantly different ($P = 0.01$)

TABLE 2. Mean length and width of colonies of three stem rust races within barley cultivars 96 hr after inoculation

Cultivar	Rust race	Colony dimensions (μm)[a]	
		Length	Width
Hiproly	Hjc	123 y	40 x
	TLM	138 y	42 x
	HDL	129 y	37 x
Larker	Hjc	69 z	26 w
	TLM	75 z	27 w
	HDL	76 z	28 w
Manker	Hjc	65 z	29 w
	TLM	71 z	29 w
	HDL	73 z	28 w

[a]Means of 40 colonies. Within columns, means followed by different letters are significantly different ($P = 0.01$)

On the same leaves used for evaluating infection types, numbers of uredia were counted. There were about nine uredia per square centimeter of leaf surface for each race on Hiproly, and about two per square centimeter on Larker and Manker (Table 1).

Development of the rust fungus.—For each race, the germination of 50-100 urediospores was observed on 30 leaves of each cultivar in two different trials. It varied from 62-71% 4 hours after inoculation and from 71-81% after 8 hours. The differences due to cultivars and to races were nonsignificant.

Appressoria of each race were observed on 30 leaves of each cultivar in two trials, 12 and 24 hours after inoculation. Appressoria associated with stomata (25 to 40 per race and cultivar) were examined and penetration was considered to have occurred from appressoria that were not stained (3). At 12 hours after inoculation penetration had occurred from 42-55% of the appressoria. These percentages changed to 61-70% 24 hours after inoculation. No significant differences could be attributed to cultivars or to races.

At 96 hours after inoculation, the length and width of the colonies of all three rust races were significantly greater within Hiproly leaf tissues than in either Larker or Manker (Table 2). Differences between Larker and Manker were not significant. Differences among races were also nonsignificant on each cultivar.

DISCUSSION

The development of *P. graminis* f. sp. *tritici* on barley cultivars has not been studied previously. We found the development of the pathogen on resistant and susceptible barley cultivars to be similar to that on resistant and susceptible wheats (8, 9, 11). There were no differences among cultivars in urediospore germination, in appressorium formation, or in penetration. However, growth of the pathogen was restricted in tissues of resistant cultivars in comparison to the susceptible cultivar. There also were fewer uredia in resistant than in susceptible cultivars. The data suggest that resistance to stem rust in barley, as conditioned by the "T" gene, is expressed after penetration has occurred and the hyphae begin to grow in the leaf tissues.

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