

Relation of Antigens of *Melampsora lini* and *Linum usitatissimum* to Resistance and Susceptibility

Abstract. A specific antigen was found in each of four races of *Melampsora lini* and four rust-differentiating varieties of *Linum usitatissimum*. Avirulence and virulence were related to resistance and susceptibility through the specific rust antigens. A race was virulent to varieties containing its specific rust antigen as a minor constituent, and avirulent to varieties lacking that antigen. No such correlation was found with flax antigens.

Numerous investigations have failed to determine the nature of the physiological or hypersensitive type of resistance to infection by rust fungi. Resistance in the host and pathogenicity in the parasite have been inherited as simple Mendelian characters in most instances (1).

The use of serologic methods as an approach to the problem is suggested by the complementary gene interaction hypothesis. This concept proposes that the mechanism governing resistance may be conditioned by a host-parasite interaction somewhat similar to an antigen-antibody reaction (2). Serology has been used to rank wheat varieties for rust resistance (3) and in studies of pollen incompatibility substances (4). It is significant that immunogenetics points to a direct relation of genes and antigens whereby the detection of antigens provides an immunochemical approach to the study of genes (5).

This report presents results of an investigation of antigens in rust-differentiating lines and varieties of *Linum usitatissimum* L. and in races of *Melampsora lini* (Ehrenb.) Lev.

Lines of flax, which are essentially alike except that each possesses a different rust-conditioning gene, were developed by backcrossing on the rust-susceptible variety Bison. In addition to Bison, lines selected from the seventh backcross to Bison of Cass, Koto, and Ottawa 770B were studied.

Two pairs of races of *Melampsora lini* having the widest differences in pathogenicity were used. The pathogenicity genotype of the urediniospore culture of each race was established by selfing. Race 22 is virulent on 23 of 25 differentials that are monogenic for rust reaction. Avirulence on Ottawa 770B differentiates race 19 from race 22. Race 1 is virulent on 4 of the 25 differentials. Virulence on Koto differentiates race 210 from race 1.

Resistance and avirulence are dominant in the lines of the host and in the races of the parasite used in this study. The reactions of each line of flax to the four races are shown in Table 1.

Globulins were prepared from the urediniospores of each race of rust and from each line of flax for use as antigens. The urediniospores were produced in the greenhouse on susceptible hosts, germinated in tap water for 6 hours at 15°C, and ball-milled with 3 percent salt solution. The suspensions were centrifuged to remove cellular debris and the supernatants were treated with 60 percent acetone to precipitate the globulins, which were frozen and dried under vacuum.

The flax globulins were prepared from greenhouse-grown plants in the prebloom stage. The plants were homogenized in a Waring blender with 3 percent salt solution. Plant and cellular debris were removed by centrifugation, and the globulins were precipitated from solution with 60 percent acetone, then frozen and dried under vacuum.

The globulins of each variety and race were reconstituted in physiological saline and used as antigens in the preparation of specific rabbit antisera. Precipitin titers for each antiserum were determined by the antiserum dilution method of Martin (6). Analyses for the various antigens present in each globulin were then carried out with each specific antiserum by precipitation reactions involving both cross-titrations and absorption tests.

The cross-titrations gave specific homologous titers of 1:2560 for each antiserum, and heterologous titers of 1:20 to 1:320, which showed that each globulin was characterized by a specific major antigen. The absorption tests demonstrated that each globulin differentially contained some of the other antigens as heterologous minor constituents which constituted a characteristic mosaic. Thus, each globulin contained not only its specific antigen, but also certain of the rust antigens as well as certain of the flax antigens.

Resistance and susceptibility were correlated to the titers of rust antiserum with flax antigens. As shown in Table 1, host-parasite interaction was susceptible when titers of rust antiserum against flax antigens were 1:160 or 1:320 and resistant when the titers were 1:20 or 1:40.

The relation of the specific rust antigens to resistance and susceptibility was more clearly shown by the absorption tests. In nine out of ten host-parasite combinations, flax varieties having a specific rust antigen as a minor constituent were susceptible to races characterized by that antigen. In all six host-parasite combinations flax varieties lacking a rust antigen as a minor constituent were resistant to races characterized by that antigen.

The absorption data suggest that in

Table 1. Association of rust reaction (R, resistant; S, susceptible) with rust antiserum titers against flax antigens. The flax varieties are shown in italic type.

<i>M. lini</i> , race 1	<i>M. lini</i> , race 19	<i>M. lini</i> , race 22	<i>M. lini</i> , race 210
<i>Cass</i> × <i>Bison</i> 7			
R 1:40	S 1:320	S 1:160	R 1:20
<i>Koto</i> × <i>Bison</i> 7			
R 1:40	S 1:320	S 1:320	S 1:320
<i>Ottawa 770B</i> × <i>Bison</i> 7			
R 1:40	R 1:40	S 1:320*	R 1:20
<i>Bison</i>			
S 1:320	S 1:320	S 1:160	S 1:320

* No antigen detected in absorption tests.

the cross-precipitation tests the association of titer differences to resistance and susceptibility is due to the presence or absence of the rust antigens in the varieties or lines of the host. The presence of a rust antigen in a flax variety would account for heterologous rust antiserum titers of 1:160 and 1:320 and the absence of the rust antigen would account for titers of 1:20 and 1:40.

Positive correlations were observed only with the rust antigens and rust antisera. No correlations were observed for the characteristic flax antigens or flax antisera, either for reaction or pathogenicity.

The specific globulin antigen of each race of rust was associated with the virulence of that race. On the other hand, the presence or absence of the rust antigens in the flax varieties was associated with the resistance or susceptibility of the variety (7).

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References and Notes

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