BREEDING FOR RESISTANCE TO RUST AND VERTICILLIUM

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The following discussion on breeding for disease resistance in sunflowers is concerned with the two main enemies of this crop in Manitoba rust and Verticillium wilt. On rust, resistance found in wild Helianthus spp. and a newly identified resistance gene are topics of discussion. In Verticillium wilt, items are presented dealing with quantitative and qualitative types of inheritance, resistance in Russian material, screening for resistance under greenhouse conditions, and effects of some chemicals as they relate to this disease.

A common and efficient source of resistance to disease affecting cultivated plants is often found in their naturally occurring wild ancestors. Sackston and Putt found resistance to rust in wild sunflower from Renner, Texas. Wild sunflowers from other locations also possess rust resistance Seed samples of wild plants were collected at locations in Canada and in the U. S. The original wild material, progenies from crosses of it with S-37-388, a highly susceptible line, and backcrosses with S-37-388 as the recurrent parent were found to possess resistance to rust in the field or under controlled conditions. Thus, resistance was found in wild sunflowers originating from Manitoba and Saskatchewan in Canada, and from the states of North Dakota, South Dakota, Kansas, Colorado, New Mexico, Oklahoma and Texas in the U.S. The species of Helianthus possessing rust resistance were mostly H. annuus but H. argophyllus from Argentina, H. petiolaris from Oklahoma and an unidentified species from New Mexico also were sources. The identity of the resistance in wild material has not been determined but some material, notably from Manitoba, North Dakota, Kansas and Oklahoma has resistance to race 3, attacking line 69-17- or the source 22 as Sackston calls it.

Presently identified rust resistance genes are R_1 and R_2 which are non-allelic and which occur, respectively, in sources 22 and 88, or respectively, in the Morden lines 69-17 and 29-3. From studies using F_1, backcross and F_2 material of progenies of crosses between lines 403-$\overline{4}$ is non-allelic and different from that of 69-17. Other unpublished studies using pure lines show that the resistance of 403-$\overline{4}$ also differs from that of 29-3. Thus a third resistance gene likely occurs in 403-$\overline{4}$ but final demonstration should come from studies using progenies of 403-$\overline{4}$ x 29-3.

Verticillium wilt of sunflowers is our most serious problem in Manitoba and breeding for resistance is the most economical and efficient means of control. A variety of resistant breeding material, falling into two groups, is available. In the one group resistance is inherited qualitatively, in the other quantitatively. Qualitative resistance is superior to quantitative resistance, at least to the Verticillium strain in Manitoba.

In qualitative inheritance, resistance is dominant and governed by the single gene V, first shown to be present in inbred line CM 144. Similar resistance has now also been found in line CM 204, unrelated to CM 144.

Quantitative inheritance of resistance to Verticillium wilt is found in a number of lines. We have been working mostly with CM 41, CM 54, and CM 214. In crosses with susceptible parents the F_1 is intermediate in resistance and is slanted slightly towards the susceptible parent. Resistant plants are recovered in F_2 populations of as low as 40 plants and not more than three and possibly only one or two genes are thought to be operative here.

Varied Russian material has yielded lines of high resistance. We have selected resistant lines out of Armavirski 9343, Smena, VNIIMK 8931 and Peredovik. Some lines have resistance as good as that of CM 144 and CM 204 previously mentioned. CM 144 was developed from the variety Mennonite and likely its resistance is also of Russian origin.

Certain kinds of resistance to Verticillium wilt can be recognized under controlled conditions in a greenhouse or growth cabinet operating at 70°F, a photoperiod of 18 hours and a light intensity of 800-1200 f.c. The resistance gene V is readily recovered when segregating F_2 progenies of crosses involving CM 144 are inoculated at the seedling stage by root immersion or stem injection using a conidial suspension. F_2 progenies segregated in a ratio of 3-Resistant : 1-Susceptible. Selfed progenies of resistant plants thus selected have been planted this year in the field, and confirm the soundness of the technique, in that resistant plants are recovered in all progenies and that progenies breeding pure and others segregating for resistance occur. The qualitative type of resistance is easily recognized under a variety of environmental conditions of temperature, light and mode of inoculation, but quantitative types of resistance are sensitive to environment and may not be expressed under certain conditions. For example, CM 54 has good field resistance but when inoculated by root immersion technique, resistance is not expressed. Inoculated by stem injection, in the greenhouse under winter conditions, CM 54 does show resistance, but not when similarly inoculated in the greenhouse during the summer.

High susceptibility to Verticillium wilt, as found in lines CM 27, CM 162 and CM 224 shows, irrespective of inoculation technique and environmental conditions. Medium susceptibility, however, as found in Advent is recognized when inoculated by root immersion but may not be recognized when inoculated by stem injection.

The last item deals with the effects of gibberellic acid and benzyladenine on Verticillium disease, and of benzyladenine on non-inoculated plants of CM 144 and CM 162, highly resistant and very susceptible, respectively, to Verticillium wilt. Gibberellic acid promoted disease in CM 162.

Under the particular experimental conditions benzyladenine had little or no effect in inoculated plants of CM 162. In non-inoculated plants of CM 162 it caused stunt, the degree increasing with increase in concentration.

At 100 ppm it caused about 60% stunt, and the general disease syndrome of stunt and leaf chlorosis was similar to that caused by the wilt organism.

In a later experiment plants of CM 144 and CM 162 were treated with a range of concentrations of benzyladenine. At 200 ppm both lines were killed quickly. At 100 ppm sharp differences were shown by the two lines: CM 162 suffered severe stunt and kill while CM 144 suffered less stunt and largely escaped injury of chlorosis and necrosis. At lower concentrations of 1 and 10 ppm CM 144 was not or only slightly stunted while CM 162 was pronouncedly stunted and about 50% of the plants developed wilt in the top portion of the plant and eventually died.

DISCUSSION

Kinman: At what temperatures did you inoculate in your Verticillium studies?

Hoes: The plants inoculated by stem injection were handled during the winter season in the greenhouse. The others, through the roots were in the growth chambers at about 70°F, 18-hour day, about 1000-foot candle of light intensity.

Sackston: How long did you keep your plants after inoculation before you felt that you could safely evaluate them? What was the period of incubation after inoculation before taking the final reading?

Hoes: I believe the stem injection method, as I used it is a bit quicker. Now I must point out that during the winter that the maximum temperatures were certainly higher than my constant temperature which I have in the growth cabinets, that may be effective. I cannot say that the particular method of inoculation per se will induce quicker symptoms. On one hand when you inoculate through the roots, the roots themselves will be affected and, of ourse, will decrease resistance to the disease process. On the other hand when you inoculate at the base of the stem the fungus does not have to affect through the roots; it is already beyond them. Again when inoculated through the stem it will not work its way down or very slowly so. It is the stem portion which goes first and the roots still may be in very good shape when inoculated through the stem.

Sackston: The reason I ask about the time is that we use the root dip inoculation technique of seedlings routinely and have done for 10 years and find, depending on conditions that sometimes we can begin to see symptoms quite clearly in as little as 7 days which is most unusual. I expect them routinely in the greenhouse, not under closely controlled conditions, in two weeks. Whenever I plan a lab for the stundents to show them the symptoms in two weeks it usually takes longer but we can generally get them in 3-3½ weeks and get our definite rating of infections. You have indicated the factors that influence the time but you haven't actually stated how long you take after inoculation to get results, days or weeks.

Hoes: It takes from 2-4 weeks by stem injection to be very certain that plants which are susceptible have succumbed and do show symptoms. It takes a little bit longer in the growth cabinets where they were inoculated by the root immersion.

Sackston: Under the conditions you are using it is a little bit slower apparently. With respect to gibberellic acid have you found the same sort of thing with it as I did if you applied it soon enough after inoculation? You could counteract the stunting effect you get from Verticillium but it wouldn't affect the total susceptiblity of the plant. By judicious treatment we could definitely avoid the stunting and yet have as much or more infection and a more drastic effect on the roots than in the presence of Verticillium alone. Of course gibberellic acid alone in appropriate concentrations has an adverse effect on the roots. What we could do was get rid of some of the symptoms but in no way could we modify, unless we aggravated it, the susceptibility of the plant. We could not make it resistant. Did you find that you could negate the stunting symptom by carefully timing gibberellic?

Hoes: No, not really.

Sackston: It is an irreversible thing if you let it go too long.

Hoes: I applied it once a week. There were no beneficial symptoms at all from gibberellic acid.

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