

BREEDING PEAS FOR RESISTANCE TO SOIL-BORNE DISEASES

Kraft, John M.

USDA/Agricultural Research Service
Washington State University
Irrigated Agriculture Research and Extension Center
Prosser, WA 99350, USA

The significant yield declines due to pea diseases prompted the pea industry in the Pacific Northwest to lobby for an ARS Plant Pathologist/Breeder position to be established at Prosser, WA. In 1964, a position was established to control soil-borne diseases of peas through breeding, chemical, cultural, and biological means.

In 1966, a pea disease nursery was established where peas are grown in monoculture. A winter wheat crop succeeds the pea crop on alternate years. This nursery is now heavily infested with several soil-borne root pathogens of peas. Soil fertilization is applied once every five years, based on recommendations of a commercial testing laboratory. The normal annual rainfall of approximately 230 mm is supplemented by sprinkler irrigation applied in 4 h sets every 7 days at ca. 13 mm of water/week. The low annual rainfall, warm temperatures, and sandy loam soil, which dries out quickly, make this field location ideal for screening candidate lines for root diseases (Fusarium, Pythium, Aphanomyces root rot, Fusarium wilt race 2), heat and drought stress.

For all field plot studies we use a fluted cone planter (ALMCO), with John Deere Max-Emerge double disc openers, which plants single, double, or 6-row plots. Seed for all plots is counted using an electronic seed counter and is treated with Captan prior to planting. Candidate, early generation lines (F3-F5) are evaluated in 3 m single plot rows with 70-100 seeds/row. Rows are planted on 112 cm centers. In alternate rows, every third plot is seeded to the control (Dark Skin Perfection = DSP). Single row plots are only compared with the nearest control plot because of the variation in soil-borne disease pressure as influenced by environment and inoculum level (10). A candidate line is considered resistant when above-ground root disease symptoms (stunting, yellowed, or dead lower leaves) are absent at flat pod stage and the yield is at least 30 % more than DSP. When stunting occurs, but above-ground symptoms are not evident, several plants within the test row are uprooted. The more resistance to uprooting, the less root disease. Single plant selections are made in heterozygous lines segregating for resistance and phenotype. It is important not to select individual plants on the ends of rows or to compare one plot with another that differs significantly in maturity. A new commercial seed stock of DSP is used each year. More advanced lines (F5-F9) are planted in 4.6 m, double row plots with 150 seeds/row and 28 cm between rows. All 2-row plots and DSP are replicated three and six times, respectively. Advanced lines (F7-F9) with desirable horticultural type are planted in 6-row plots, 6.1 m long. Two hundred seeds/row are planted and one-half of each of three replicates is harvested for freezing and taste evaluation tests; the remainder is dried and harvested for seed. All 6-row plots, including DSP, are replicated four times.

In the winter months, crossing and disease screening are conducted in the greenhouse and growth chamber. A crossing program is conducted where the most resistant USDA lines, germplasm releases from other institutions and selections from tolerant commercial lines, are intercrossed and the

screening and evaluation repeated.

Candidate lines are planted into soil bins (1.5 m x 1.5 m x 0.3 m high) filled with soil artificially infested with Fusarium solani (Mart.) Sacc. f. sp. pisi (Jones) Snyd. & Hans., Pythium ultimum Trow, and Aphanomyces euteiches Drechs. Supplemental light (4 h illumination) is created by metal halide lamps which produce 1000 ft/c at the soil surface. Greenhouse temperatures vary from 20-25°C. Twenty seeds are planted per line. Four replications of DSP are planted in each of four bins. Test plants are watered when the top 2 cm of soil is dry. Candidate lines are evaluated for resistance at 3-4 weeks after emergence by carefully digging, washing the roots, and scoring each plant on a 0-5 scale, where 0 = healthy root and 5 = a completely rotted root. Plants obviously segregating for resistance (score a 0-2) in a heterozygous line are transplanted and saved for seed. Those lines which outyielded DSP by 30 % in the field nursery and scored a 0-2 in the greenhouse test are considered resistant to Fusarium, Pythium, and Aphanomyces root rot.

Specific resistance to Fusarium root rot, caused by Fusarium solani f. sp. pisi, is determined in artificially infested soil (40,000 colony forming units/g air dry soil) in a controlled environment (1). Resistant plants (a 0-2 score) from segregating populations are saved and grown to produce seed, are increased and reevaluated. Our resistant and susceptible check varieties for Fusarium root rot are P.I. 257593 and DSP (1), respectively.

Resistance/tolerance to Aphanomyces root rot is determined in the greenhouse at 22.25 °C. Test lines, 20 seeds/line, are planted in flats of coarse-grade perlite and 10-day-old seedlings are inoculated with a zoospore concentration of 3-8 x 10⁵/ml. Zoospore inoculum of A. euteiches is produced by incubating the fungus in autoclaved pea seed broth (10 g pea seed/100 ml distilled water) for 7 days in the dark at 22°C. The resultant mycelial mats are washed in sterile tap water and incubated overnight on the laboratory bench in a minimal salt solution with filtered air bubbling in each flask (7 mats/300 ml solution). The resultant zoospore inoculum concentration is determined with a haemocytometer. Plants are inoculated by removing them from perlite and dipping the roots in the zoospore suspension. Inoculated plants are transplanted back into the perlite and compared with the non-inoculated controls after a 2-week incubation. Any lines which produce at least 70 % of the fresh plant weight of control plants of the same line are considered resistant. With the exception of a few highly resistant P.I. lines, most inoculated lines will score a 4-5 using the root disease severity scale. Therefore, fresh weights of test plants are used to determine resistance or susceptibility. As an example, DSP will score a 4-5 disease severity rating but will average only 40 % fresh plant weight of the uninoculated control. Our resistant check for A. euteiches is P.I. 180693, which will exhibit a

disease severity rating of 0-2 with most isolates of A. euteiches used and will produce 80 % fresh weight of the uninoculated control. Candidate lines which survive the greenhouse test are also evaluated in the field where A. euteiches is severe. Dr. David Davis, Univ. of Minnesota, St. Paul, MN, Dr. Ted Reiling, Green Giant/Pillsbury, Le Sueur, MN, Drs. Don Hagedorn and Earl Gritton at the Univ. of Wisconsin, Madison, WI, have cooperated with us the last several years.

Resistance to Fusarium wilt caused by Fusarium oxysporum Schl. f. sp. pisi (van Hall) Snyd. & Hans, race 1 (Snyd. & Hans) is determined under

field conditions at Pullman, WA, in cooperation with Dr. Fred Muehlbauer, who has established a race 1 nursery. Resistance to race 2 Snyder & Hans., race 5 Hagl. and Kraft, and race 6 Hagl. and Kraft, is determined in pure culture in the greenhouse, also in coarse grade perlite by procedures described previously (8). The differential varieties and germplasm lines used and their sources are listed in Table 1.

Table 1. Fusarium wilt differential varieties

Varieties and germplasm used	Seed source	Wilt reaction			
		R1	R2	R5	R6
M410	Brotherton Seed Co.	S*	S	S	S
Vantage	Brotherton Seed Co.	R	S	S	S
Mini	Asgrow Seed Co.	S	R	S	S
Mini 93	Asgrow Seed Co.	R	R	S	S
Sundance II	Pure Line Seed Co.	R	S	R	S
Grant	Brotherton Seed Co.	R	S	S	R
74SN5	USDA/ARS	R	R	R	R

* S = susceptible reaction, R = resistant reaction

A pedigree-recurrent selection program is used and was computerized by adapting the MSTAT version-2 program developed and made available by Michigan State University. Selection for various disease and stress resistance is delayed until the F4 because of the usual presence of non-additive variance in divergent crosses. Approximately 2500 separate lines are identified by hybrid number for: 1) generation; 2) whether bulked or selected; 3) horticultural and disease resistance attributes; and 4) seed characteristics. The long range goal of this program is to develop multiple disease resistant/tolerant germplasm lines in divergent backgrounds. These lines must approach commercial type and be only a few crosses away from commercial use. It appears that root disease resistance, excluding Fusarium wilt, is quantitatively inherited and outcrossing with susceptible parents greatly reduces the chances of recovering resistant progeny.

We have officially released 20 breeding lines since 1971 which are available upon request (2,3,4,5,6,9,11). We have an open door policy and advocate the free exchange of germplasm. We only ask that this be mutual. We encourage visits to our nursery during the growing season (June 20-July 14).

1. Kraft, J.M. 1975. Plant Dis. Reprtr. 59:1007-1011.
2. Kraft, J.M. 1981. Crop Sci. 21:352-353.
3. Kraft, J.M. 1984. Crop Sci. 24:389.
4. Kraft, J.M. 1988. PNL 20:58.
5. Kraft, J.M. and R.A. Giles. 1976. Crop Sci. 16:126.
6. Kraft, J.M. and R.A. Giles. 1978. Crop Sci. 18:1098.

7. Kraft, J.M. and R.A. Giles. 1978. Crop Sci. 18:1099.
8. Kraft, J.M. and W.A. Haglund. 1978. Phytopathology 68:273-275.
9. Kraft, J.M. and J.A. Tuck. 1986. Crop Sci. 26:1262-1263.
10. Kraft, J.M., D.W. Burke and W.A. Haglund. 1981. In: Fusarium Diseases: Biology and Taxonomy, Penn. State Univ. Press, eds. P.E. Nelson, T.A. Toussoun, and R.J. Cook. Pp. 157-168.
11. Kraft, J.M., M.J. Silbernagel and F.J. Muehlbauer. 1972. Crop Sci. 12:399.
