## Distribution of Heterodera glycines in Ohio<sup>1</sup>

H. R. WILLSON,<sup>2</sup> R. M. RIEDEL,<sup>3</sup> J. B. EISLEY,<sup>4</sup> C. E. YOUNG,<sup>5</sup> J. R. JASINSKI,<sup>6</sup> T. A. WHEELER,<sup>7</sup> P. H. KAUFFMAN,<sup>8</sup> P. E. PIERSON,<sup>9</sup> AND M. C. STUART<sup>9</sup>

Abstract: A 4-year systematic survey for the presence of soybean cyst nematode Heterodera glycines in Ohio soybean fields was initiated in 1992. A total of 667 soybean fields in 63 counties was sampled. Heterodera glycines was present in 91 fields in 40 counties based on soil samples collected, and in one field in each of three additional counties based on soil samples submitted to the Plant and Pest Diagnostic Clinic or through a preliminary survey conducted in 1991. Soybean hectarage in the 43 counties with at least one field known to be infested with H. glycines accounts for 79% of the total Ohio soybean production area. Eight races of H. glycines were identified in 33 samples from 18 counties. The most common was race 3, identified in 15 samples; others were races 1, 2, 4, 5, 6, 10, and 14.

Key words: distribution, Glycine max, Heterodera glycines, Ohio, soybean, soybean cyst nematode, survey, races.

In 1994, Ohio ranked fifth in soybean production in the United States. Ohio produces soybeans on approximately 1.6 million hectares annually. In 1994, crop value was estimated at \$990 million. Soybean cyst nematode *Heterodera glycines* was first detected in Miami County, Ohio, in 1980 in soil accompanying tomato transplants shipped from Tennessee (3). In 1987, soybean field infestations were detected in Sandusky County in northern Ohio and Scioto County in southern Ohio (9). In 1991, a survey for the nematode was conducted by the Ohio Cooperative Agricul-

<sup>4</sup> Research Associate, Department of Entomology, The Ohio State University, 1991 Kenny Road, Columbus, OH 43210. ture Pest Survey program. Seventy fields distributed among 33 counties were sampled. The nematode was detected in Darke, Erie, Fulton, Huron, Preble, Seneca, Union, and Wood counties. A systematic survey was initiated in 1992 to determine the distribution of *H. glycines* in Ohio soybean fields.

## MATERIALS AND METHODS

The survey was initiated with a general target of sampling 10 soybean fields in each Ohio county with 2,000 or more hectares of soybean production. Sampling preference was given to fields that were in the second year of soybean production, or those suspected by growers to have a production problem. Fields were located with the cooperation of county extension agents. A total of 667 fields in 63 counties were sampled during the growing seasons of 1992 through 1995. Field sampling included a preliminary visual inspection of soybean root systems for the presence of cysts and the collection of a composite soil sample (ca. 1 liter), which was generally obtained from the field entrance and areas exhibiting symptoms of plant stress, or obtained from random sites within a field. Soil samples were transported in insulated chests to the laboratory and stored at 10 °C until processed.

All soil samples were bioassayed to de-

Received for publication 15 April 1966.

<sup>&</sup>lt;sup>1</sup> Research supported in part by a grant from the Ohio Soybean Council. Paper No. 47-96 of the Ohio Agricultural Research and Development Center.

<sup>&</sup>lt;sup>2</sup> Associate Professor, Department of Entomology, The Ohio State University, 1991 Kenny Road, Columbus, OH 43210.

<sup>&</sup>lt;sup>3</sup> Professor, Department of Plant Pathology, The Ohio State University, 2021 Coffey Road, Columbus, OH 43210.

<sup>&</sup>lt;sup>5</sup> Extension Associate, Northwest Extension District, 952 Lima Avenue, Findlay, OH 45840.

 <sup>&</sup>lt;sup>6</sup> Extension Associate, Southwest Extension District, 303
Corporate Center Drive, Vandalia, OH 45377.
<sup>7</sup> Former Post-Doctoral Researcher, Department of Plant

<sup>&</sup>lt;sup>7</sup> Former Post-Doctoral Researcher, Department of Plant Pathology, The Ohio State University, 2021 Coffey Road, Columbus, OH 43210. Currently Assistant Professor, Texas Agricultural Experiment Station, Rt. 3, Box 219, Lubbock, TX 79401.

<sup>&</sup>lt;sup>8</sup> Plant Pest Division, Ohio Department of Agriculture, Reynoldsburg, OH 43068.

<sup>&</sup>lt;sup>9</sup> Post-Doctoral Researchers, Department of Plant Pathology, The Ohio State University, 2021 Coffey Road, Columbus, OH 43210.

The contributions of Z. Chen, R. Lyons, and F. Hale to this research are gratefully acknowledged.

E-mail: willson.1@osu.edu

termine the presence of *H. glycines.* Three seeds of the susceptible soybean cultivar Corsoy 79 were planted in 350 cm<sup>3</sup> soil in a 10-cm-diam. plastic pot. Seedlings were thinned to one per pot after emergence. Plants, grown at 24 °C, were watered, fertilized, and treated for insect infestations as needed. After 5 weeks cysts were extracted. Soil was suspended in water in a

10-liter bucket and passed through stacked sieves (840- $\mu$ m-pore over 250- $\mu$ m-pore). Roots were washed over the sieves to recover cysts. Material collected on the 250- $\mu$ m-pore sieve was inspected for the presence of new cysts using a dissecting microscope. Sites were declared positive if the bioassay produced cysts.

In addition, 200-cm<sup>3</sup> soil samples were

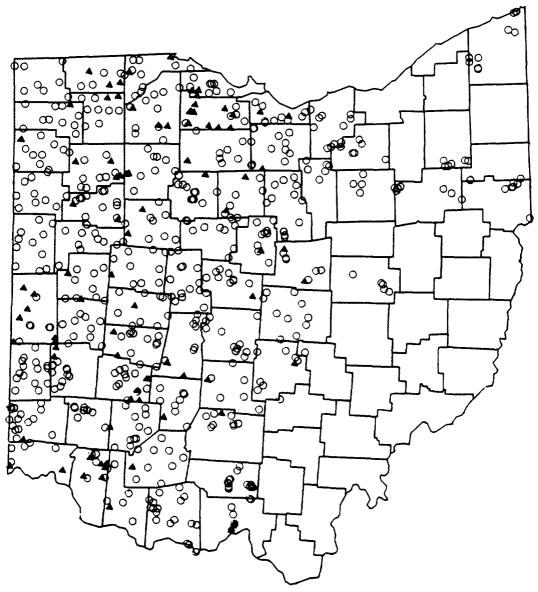


FIG. 1. Sites sampled in a detection survey for *Heterodera glycines* in soybeans in Ohio from 1992 through 1995. ( $\blacktriangle$  positive,  $\bigcirc$  negative)

	Index <sup>a</sup>				
County	Pickett	Peking	PI 88788	PI 90763	Race
Hardin	$1.8 \pm 0.7$	0	$15.8 \pm 3.2$	0	1
Seneca	$0.2 \pm 0.1$	$0.2 \pm 0.1$	$32.5 \pm 4.2$	$0.1 \pm 0.1$	1
Greene	$81.9 \pm 13.8$	$17.3 \pm 5.2$	$16.2 \pm 5.3$	$1.6 \pm 0.8$	2
Putnam	$30.6 \pm 5.0$	$21.1 \pm 4.1$	$11.2 \pm 3.5$	$8.1 \pm 2.4$	2
Sandusky	$61.2 \pm 15.4$	$21.3 \pm 7.0$	$54.5 \pm 8.2$	$7.0 \pm 2.0$	2
Clermont	$5.2 \pm 1.1$	$0.1 \pm 0.1$	$0.7 \pm 0.3$	0	3
Clermont	0	$0.3 \pm 0.1$	$1.7 \pm 1.0$	0	3
Darke	0	0	$1.6 \pm 1.0$	$0.3 \pm 0.3$	3
Erie	$1.9 \pm 0.7$	$0.2 \pm 0.2$	$2.1 \pm 1.4$	0	3
Fulton	0	0	0	0	3
Fulton	$5.1 \pm 1.6$	0	$3.3 \pm 1.4$	0	3
Greene	$7.7 \pm 2.3$	$0.5 \pm 0.2$	$7.8 \pm 2.0$	$0.3 \pm 0.2$	3
Hamilton	$1.7 \pm 1.0$	$0.8 \pm 0.8$	$3.4 \pm 0.8$	0	3
Henry	$0.4 \pm 0.2$	0	$3.9 \pm 0.8$	0	3
Huron	$0.8 \pm 0.1$	$0.2 \pm 0.2$	$8.8 \pm 3.0$	0	3
Mercer	$0.2 \pm 0.2$	$0.1 \pm 0.1$	$0.7 \pm 0.2$	0	3
Miami	$1.7 \pm 0.6$	0	$6.6 \pm 1.0$	0	3
Paulding	$2.9 \pm 0.6$	$0.2 \pm 0.1$	$1.5 \pm 0.5$	0	3
Putnam	$1.0 \pm 0.4$	0	$6.1 \pm 1.9$	$1.0 \pm 0.7$	3
Wood	$1.9 \pm 0.5$	$1.0 \pm 0.1$	$6.4 \pm 2.1$	0	3
Wood	$4.2 \pm 0.5$	$1.1 \pm 0.4$	$8.9 \pm 0.9$	$1.4 \pm 0.7$	3
Wood	0	0	$7.4 \pm 1.2$	$0.9 \pm 0.6$	3
Wood	$1.1 \pm 0.6$	$0.1 \pm 0.1$	$4.2 \pm 0.4$	0	3
Ottawa	$65.2 \pm 21.7$	$23.8 \pm 4.3$	$26.1 \pm 9.5$	$10.4 \pm 4.1$	4
Sandusky	$13.7 \pm 2.0$	$3.1 \pm 0.5$	$15.6 \pm 3.8$	$2.4 \pm 0.2$	5
Scioto	$55.8 \pm 9.8$	$3.9 \pm 1.1$	$67.1 \pm 9.6$	$0.5 \pm 0.1$	5
Clermont	$107.4 \pm 34.3$	$0.4 \pm 0.2$	$1.8 \pm 0.6$	$0.1 \pm 0.1$	6
Ottawa	$14.4 \pm 0.8$	$4.0 \pm 1.1$	$2.8 \pm 1.0$	$1.4 \pm 0.7$	6
Putnam	$13.5 \pm 3.2$	0	$8.2 \pm 1.4$	$1.5 \pm 1.5$	6
Sandusky	$13.9 \pm 6.5$	$7.7 \pm 1.2$	$2.8 \pm 0.9$	$4.4 \pm 1.3$	6
Sandusky	$28.1 \pm 7.6$	$3.6 \pm 1.3$	$9.4 \pm 2.0$	$6.8 \pm 2.8$	6
Huron	$26.8 \pm 7.3$	$2.1 \pm 0.7$	$1.4 \pm 0.4$	$11.0 \pm 2.7$	10
Crawford	$98.3 \pm 25.5$	$56.4 \pm 14.2$	$4.0 \pm 1.2$	$42.0 \pm 10.4$	14

TABLE 1. Mean female indices, followed by standard error on differential soybean genotypes used for race characterization of *Heterodera glycines* populations detected in Ohio.

<sup>a</sup> Index = percentage of cysts on resistant cultivars relative to Lee.

assayed for the presence of cysts (1992) or second-stage juveniles (J2) and eggs (1993-1995). In 1992, field samples were processed by the Fenwick Can elutriator (1) followed by microscopic examination of screen material for cysts. From 1993 through 1995, soil samples were initially assayed for J2 juveniles and other plantparasitic nematodes with a modified Baermann funnel extraction method (11). Samples also were assayed for eggs by first extracting cysts from a 200-cm<sup>3</sup> soil subsample using the same technique described for the bioassays. Cysts were crushed in a 40-cm<sup>3</sup> Ten Broeck tissue grinder and the debris poured over a 25µm-pore sieve. The eggs were recovered from the sieve, stained with acid fuchsin, and counted (4).

Race determinations were conducted on *H. glycines* isolates from 33 infested fields (10). To increase an *H. glycine* population, cysts from the bioassay were crushed and the eggs were added to soil (1.5 parts construction sand:1 part loam) in 15-cm-diam. clay pots in which seedlings of Corsoy 79 were growing. Plants were grown for 30 to 60 days. For race tests, three seeds each of the susceptible Lee 74 and the differential soybean lines Pickett, Peking, PI88788, and PI90763 were planted in construction sand in 350-cm<sup>3</sup> pots. Six replicate pots of each of the soybean lines were maintained in 25.4-cm-diam.  $\times$  50.8-cm-diam. plastic

trays in a 24 °C greenhouse. In order to avoid washing out vermiform juveniles, plants were watered by the ebb-and-flow method. Week-old seedings were thinned to one plant per pot and the soil infested with 4,000 to 6,000 eggs from the isolate to be tested. After 4 to 5 weeks, cysts were washed from the roots with a highpressure water spray, extracted from the sand, and counted. A female index (FI) was calculated for each differential as follows: FI = (mean number of cysts on differential)/(mean number of cysts on Lee 74)  $\times 100$ . Race classification was assigned according to the system established by Riggs et al. (10).

## **RESULTS AND DISCUSSION**

The 63 counties sampled account for 99% of Ohio's total soybean hectarage. *H.* glycines was detected in 91 sites in 40 counties. The 40 infested counties represent 73% of the state's soybean hectarage. With two additional counties found to be positive from the 1991 survey and one from Plant and Pest Diagnostic Clinic records, soybean hectarage in positive counties increases to 79%. In general, the nematode was detected throughout western and central Ohio but not in the eastern region of the state where soybean production is less intensive (Fig. 1).

Since the emphasis of this survey was on detection and sites sampled were either in the second consecutive year of soybean production or regarded by growers as problem fields, the frequency of H. glycines infestations detected may not accurately reflect the frequency of H. glycines infestation of Ohio soybean fields. However, the widespread detection does reflect the potential risk of H. glycines infestations in this state. Given the widespread distribution of H. glycines in this survey, all the major soybean-producing counties of Ohio are subject to a potential risk of infestation.

Race determinations were completed for 33 fields sampled from 18 counties. Eight of the possible 16 races were identified (Table 1). Race 3, the most common, was found in 19 of 33 samples from 13 counties. Five of 33 populations from four counties were identified as race 6. Race 2 was identified in three counties, and races 1 and 5 were identified in two counties. Races 4, 10, and 14 were found in a single site each.

The race distribution in Ohio is similar to that reported for Missouri (7). Race 3 was the predominant race identified in both Missouri and Ohio, and six of the seven other races identified in Ohio match the six remaining races reported for Missouri. However, race 6 is the second most common race in Ohio, whereas race 1 was the second most common in Missouri. This contrasts with race distribution for Arkansas and North Carolina (10), where races 2 and 4 predominate, and for South Carolina (5), where race 14 is the most common and race 3 the second most common.

The impact of H. glycines on soybean yield production in the midwestern states has been documented (2,6,8). Field evaluations of resistant and susceptible soybean cultivars in Ohio during the past 2 years have demonstrated that yields of susceptible cultivars in infested soils are reduced an average of 15% (unpubl. data). Given such reductions in yield, H. glycines must be regarded as a significant threat to Ohio soybean production. The nematode has been detected in a major proportion of soybean-producing counties in Ohio, and infestations causing yield losses have been observed in a number of areas within the state. In addition, race determination results have demonstrated that populations of H. glycines are genetically diverse in Ohio.

## LITERATURE CITED

1. Byrd, D. W., Jr., K. R. Barker, H. Ferris, C. J. Nusbaum, W. E. Griffin, R. J. Small, and C. A. Stone. 1976. Two semiautomatic elutriators for extracting nematodes and certain fungi from soil. Journal of Nematology 8:206-212.

2. Doupnik, B., Jr. 1993. Soybean production and disease loss estimates for northcentral United States from 1989 to 1991. Plant Disease 77:1170-1171.

3. Hammond, R. B., P. H. Kauffman, R. M.

Riedel, and R. E. Hite. 1981. Soybean cyst nematode introduced into Ohio. Plant Disease 65:846.

4. Hussey, R. S. 1985. Staining nematodes in plant tissues. Pp. 197–199 in B. M. Zuckerman, W. F. Mai, and M. B. Harrison, eds. Plant nematology laboratory manual. Amherst, MA: The University of Massachusetts Agriculture Experiment Station.

5. Lewis, S. A., C. E. Drye, J. A. Saunders, E. R. Shipe, and J. M. Halbrendt. 1993. Plant-parasitic nematodes on soybean in South Carolina. Supplement to the Journal of Nematology 25:890-894.

6. Niblack, T. L., N. K. Baker, and D. C. Norton. 1992. Soybean yield losses due to *Heterodera glycines* in Iowa. Plant Disease 76:943–948.

7. Niblack, T. L., R. D. Heinz, G. S. Smith, and

P. A. Donald. 1993. Distribution, density, and diversity of *Heterodera glycines* in Missouri. Supplement to the Journal of Nematology 25:880–886.

8. Niblack, T. L., ed. 1993. Protect your soybean profits: Manage soybean cyst nematode. Columbia, MO: University of Missouri Printing Services.

9. Riedel, R. M., and A. M. Golden. 1988. First report of *Heterodera glycines* on soybean in Ohio. Plant Disease 72:363.

10. Riggs, R. D., and D. P. Schmidt. 1988. Complete characterization of the race scheme for *Heterodera glycines*. Journal of Nematology 22:392–395.

11. Thistlethwayte, B. 1970. Reproduction of *Pratylenchus penetrans* (Nematoda: Tylenchida). Journal of Nematology 2:101–105.