

DISTRIBUTION OF *HETERODERA GLYCINES* RACES IN BRAZIL

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ABSTRACT

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Soybean cyst nematode, *Heterodera glycines*, populations were obtained from the states of Goiás, Mato Grosso, Mato Grosso do Sul, and Minas Gerais in 1993 from infested fields and greenhouse cultures. Collections were brought under quarantine to Urbana, Illinois for race identification. Nematodes were cultured on 'Lee 68' soybean, and races were identified using 'Lee 68' as the susceptible host and the soybeans 'Pickett 71', 'Peking', PI88.788, and PI90.763 as differential hosts. Plants were inoculated with second-stage juveniles and after approximately 1 month, numbers of white females that had developed on each plant were determined. A female index (FI = $[\bar{x}$ of females that developed on the differential] / $[\bar{x}$ of females that developed on Lee 68] \times 100) was calculated for each differential soybean line and a race was designated for each population. Races 2, 3, and 5 were identified in collections obtained in Mato Grosso. Races 3 and 14 were identified in collections from Goiás. Races 3, 4, 10, and 14 occurred in Mato Grosso do Sul. Race 3 was the only race identified in collections obtained in Minas Gerais.

Key words: geographical distribution, *Glycine max*, *Heterodera glycines*, races, soybean, soybean cyst nematode.

RESUMEN

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Poblaciones del nematodo del quiste, *Heterodera glycines*, fueron obtenidas de los estados de Goiás, Mato Grosso, Mato Grosso do Sul, y Minas Gerais en 1993, de campos infestados y de cultivos de invernadero. Muestras de estos nematodos fueron traídas bajo cuarentena a Urbana, Illinois para la identificación de razas. Los nematodos fueron mantenidos en soya 'Lee 68' y la identificación de las razas se hizo utilizando 'Lee 68' como hospedero susceptible y soya 'Pickett 71', 'Peking', PI88.788, y PI90.763 como hospederos diferenciales. Las plantas fueron inoculadas con juveniles del segundo estadio y luego en aproximadamente un mes, se determinó el número de hembras blancas que se desarrollaron en cada planta. Se calculó un índice de hembras (IH = $[\bar{x}$ de hembras que se desarrollaron en el hospedero diferencial] / $[\bar{x}$ de hembras que se desarrollaron en 'Lee 68'] \times 100), para cada uno de los hospederos diferenciales y se designó una raza para cada población. Las razas 2, 3, y 5 fueron indentificadas de poblaciones provenientes de Mato Grosso. Las razas 3 y 4 fueron identificadas en muestras provenientes de Goiás. Las razas 3, 4, 10, y 14 ocurrieron en Mato Grosso do Sul. La raza 3 fue la única de las razas indentificada en muestras provenientes de Minas Gerais.

Palabras clave: distribución geográfica, *Glycine max*, *Heterodera glycines*, nematodo del quiste de la soya, razas, soya.

INTRODUCTION

Brazil produced soybean [*Glycine max* (L.) Merr.] on 11.4 million hectares in 1992–1993. Prior to the 1970's, soybean production was limited to the southern states of Paraná, Rio Grande do Sul, Santa Catarina, and São Paulo (Fig 1.). During the 1970's, soybean production in the central savannah began in the states of Goiás, Mato Grosso, and Mato Grosso do Sul, and the number of hectares planted increased dramatically in the 1980's. The number of hectares in the central savannah has continued to increase in the 1990's. Farms also

have been established recently in north-eastern Brazil where the borders of Bahia, Maranhão, Piauí, and Tocantins intersect. The cropping system in the central savannah and in the northeast consists primarily of monoculture soybean. During the 1991–1992 growing season, *Heterodera glycines* Ichinohe was found for the first time in Brazil in the state of Minas Gerais on a farm having a 10-year history of soybean grown in monoculture (3). Subsequent to this finding, *H. glycines* was found in the states of Goiás, Mato Grosso, and Mato Grosso do Sul (3). In 1993, samples were collected from several infested farms in



Fig. 1. States that produce soybean and locations of *Heterodera glycines* collections obtained in Brazil during 1993: Brazil 1,2 = Nova Ponte, Minas Gerais; Brazil 3,4 = Chapadão do Sul, Mato Grosso do Sul; Brazil 5,6 = Aporé, Goiás; Brazil 8 = Costa Rica, Mato Grosso do Sul; Brazil 9 = Iraí de Minas, Minas Gerais; Brazil 10,12 = Campo Verde, Mato Grosso; Brazil 11 = Jaciara, Mato Grosso; Brazil 13 = Diamantino, Mato Grosso; Brazil 14 = Tangará da Serra, Mato Grosso; Brazil 15 = Campo Novo dos Parecís, Mato Grosso; and Brazil 16 = Chapadão do Céu, Goiás.

these four states and were brought under quarantine to Urbana, Illinois for study. The objectives of the research reported herein were to identify the races of *H. glycines* present in Brazil and to determine their distribution.

MATERIALS AND METHODS

Soil samples were collected in infested fields in February, 1993 near Nova Ponte, Minas Gerais (Brazil 1 and Brazil 2), Chapadão do Sul, Mato Grosso do Sul (Brazil 3 and Brazil 4), Aporé, Goiás (Brazil 5 and Brazil 6), and 20 km east of Costa Rica, Mato Grosso do Sul (Brazil 7 and Brazil 8) (Fig. 1). Two additional samples were obtained from collections maintained on 'Cristalina' soybean in a greenhouse at the National Soybean Research Center, Londrina, Paraná. These samples originated from Iraí de Minas, Minas Gerais (Brazil 9) and from Campo Verde, Mato Grosso (Brazil 10). In August, 1993, additional greenhouse samples maintained on 'Cristalina' were obtained from collections made at different times during the latter portion of the 1992–1993 growing season. These samples originated from Jaciara, Mato Grosso (Brazil 11), Campo Verde, Mato Grosso (Brazil 12), Diamantino, Mato Grosso (Brazil 13), Tangará da Serra, Mato Grosso (Brazil 14), Campo Novo dos Parecis, Mato Grosso (Brazil 15), and Chapadão do Céu, Goiás (Brazil 16).

Approximately 300 cm³ of soil from each sample was mixed with an equal volume of sand and placed in 700-cm³ pots on a greenhouse bench in the USDA Nematology Greenhouse located on the University of Illinois campus. Nematodes were cultured on 'Lee 68' soybean until sufficient numbers of second-stage juveniles (J2) could be obtained for experimentation. Seeds of 'Lee 68', 'Pickett 71', 'Peking', PI88.788, and PI90.763 were ger-

minated, and when radicles were 3–4 cm long, seedlings were planted individually in 300-cm³ pots containing 250 cm³ of an autoclaved 3:1 sand:sandy loam growing medium. Nematode inoculum was prepared by extracting cysts from soil using gravity-sieving with nested 850 and 250- μ m-pore sieves. Cysts and debris were poured into a counting dish, and cysts were removed individually with forceps, crushed in a tissue grinder, and the resulting suspension poured onto a modified Baermann funnel with the screen placed in distilled deionized water. After 2–3 days J2 were collected, counted, and the nematode suspension pipeted in equal volumes into three equidistant, 3-cm-deep holes that formed the points of a triangle around the soybean seedlings. Depending on the numbers of J2 obtained for each population, 500, 800, or 1 000 J2 were added to each pot approximately 1 week after transplanting seedlings. Each soybean line was replicated five times in a completely randomized design. After approximately 1 month, numbers of white females that developed on each plant were determined and a female index (FI) was calculated for each differential soybean (7) where $FI = [(\bar{x} \text{ of females that developed on the differential}) / (\bar{x} \text{ of females that developed on 'Lee 68'})] \times 100$. A race designation (6) was assigned based on a +/- system where $FI \geq 10\% = +$ and $FI < 10\% = -$.

RESULTS AND DISCUSSION

Races 2, 3, 4, 5, 10, and 14 were identified (Table 1), and the locations are indicated in Fig. 1. Race 2 occurred only in Mato Grosso (Brazil 11). Race 3 was identified most frequently and occurred in Minas Gerais (Brazil 1, Brazil 2, and Brazil 9), Goiás (Brazil 6 and Brazil 16), and Mato Grosso (Brazil 12–15). Race 3 was the only race that occurred in Minas Gerais.

Brazil 1 was collected from the field where *H. glycines* was first found in Brazil. Race 4 was found once in a varietal trial in Mato Grosso do Sul (Brazil 3). Race 5 also was identified once and was found in Mato Grosso (Brazil 10). The race 10 population was found once (Brazil 8) about 20 km east of Costa Rica and 50 km west of Chapadão do Sul, Mato Grosso do Sul where collections Brazil 3 and Brazil 4 were obtained. Race 14 was found twice (Brazil 4 and Brazil 5). Collection Brazil 4 was obtained from the rhizosphere of a resistant cultivar in the same field experiment as collection Brazil 3. Collections Brazil 5 and Brazil 6 were obtained from the same field about 300 m apart in Aporé County, Goiás. Although collections Brazil 5 and Brazil 6 were obtained in Goiás, they were taken on the same farm as collections Brazil 3 and

Brazil 4. The field plot and the production field were about 10 km apart. Brazil 6 was from an area of the field in which the soybeans were severely chlorotic and expressed symptoms typical of infection due to race 1 in North Carolina, U.S.A. (1,8). Symptoms probably were due to aluminum toxicity resulting from low soil pH. Race 1, which is common in China and Japan (Noel, unpublished) and the midwestern (4,9) but not the southern U.S.A. (2,10), was not identified in any of the collections obtained in Brazil. Race 6, which comprised 5–15% of races identified in some surveys in the U.S.A. (2,4,10), was not identified in any of the Brazilian populations. Nematodes obtained in collection Brazil 7 did not reproduce under greenhouse conditions.

Table 1. Distribution and identification of *Heterodera glycines* races in Brazil.

| Collection | State | Female index (FI) on differential soybean lines (%) ^y | | | | Race ^z |
|------------|--------------------|--|----------|----------|----------|-------------------|
| | | Pickett 71 | Peking | PI88.788 | PI90.763 | |
| Brazil 1 | Minas Gerais | 1.9 (-) | 0.0 (-) | 0.0 (-) | 0.0 (-) | 3 |
| Brazil 2 | Minas Gerais | 0.2 (-) | 0.0 (-) | 0.1 (-) | 0.0 (-) | 3 |
| Brazil 3 | Mato Grosso do Sul | 38.6 (+) | 13.3 (+) | 12.2 (+) | 12.2 (+) | 4 |
| Brazil 4 | Mato Grosso do Sul | 92.9 (+) | 26.8 (+) | 3.9 (-) | 34.9 (+) | 14 |
| Brazil 5 | Goiás | 98.3 (+) | 31.5 (+) | 3.5 (-) | 18.2 (+) | 14 |
| Brazil 6 | Goiás | 6.4 (-) | 2.5 (-) | 1.4 (-) | 2.8 (-) | 3 |
| Brazil 8 | Mato Grosso do Sul | 93.2 (+) | 9.8 (-) | 0.7 (-) | 39.6 (+) | 10 |
| Brazil 9 | Minas Gerais | 0.0 (-) | 0.0 (-) | 2.0 (-) | 0.0 (-) | 3 |
| Brazil 10 | Mato Grosso | 43.2 (+) | 8.8 (-) | 47.1 (+) | 0.2 (+) | 5 |
| Brazil 11 | Mato Grosso | 82.5 (+) | 28.7 (+) | 43.1 (+) | 0.0 (-) | 2 |
| Brazil 12 | Mato Grosso | 6.3 (-) | 0.0 (-) | 0.0 (-) | 0.0 (-) | 3 |
| Brazil 13 | Mato Grosso | 0.0 (-) | 0.0 (-) | 0.0 (-) | 0.0 (-) | 3 |
| Brazil 14 | Mato Grosso | 0.0 (-) | 0.0 (-) | 0.1 (-) | 0.1 (-) | 3 |
| Brazil 15 | Mato Grosso | 0.0 (-) | 0.0 (-) | 0.0 (-) | 0.0 (-) | 3 |
| Brazil 16 | Goiás | 0.0 (-) | 0.0 (-) | 0.0 (-) | 1.8 (-) | 3 |

^yFI = (\bar{x} of females that developed on the differential) / (\bar{x} of females that developed on 'Lee 68') × 100.

^zRace designation (6) based on a +/- system where FI ≥ 10% = + and FI < 10% = -.

The means and date of introduction of *H. glycines* into Brazil is not known. The nematode may have a history analagous to that of its history in the U.S.A. (5). The nematode may have been introduced more than once. Many Brazilian agriculturists travel to the U.S.A., and many Brazilians have Japanese ancestry and travel to Japan. Ample opportunity has existed for introduction of the nematode from these two countries. There is no evidence of used farming equipment being moved from either the U.S.A. or Japan to Brazil. Although unlikely, *H. glycines* may have been imported from Colombia, the only other South American country with documented infestations of *H. glycines*. Recency of soybean farming in the savannah of Brazil and widespread distribution of the nematode suggest that *H. glycines* was moved with seed lots from the seed production area in Minas Gerais. The distribution and frequency of occurrence of race 3 in Minas Gerais and the other three states support this hypothesis. Races other than race 3 may have been introduced separately or may have developed due to selection pressure other than planting of resistant cultivars. Two cycles of planting resistant cultivars may have caused the selection of the race 4 and race 14 populations in the research plot from which Brazil 3 and Brazil 4 were obtained (Table 1). Occurrence of different races in the same field (Brazil 5 and Brazil 6) is not unexpected given the variability of *H. glycines* and propensity for localized inbreeding due to the aggregated distribution and relative lack of movement of *H. glycines* within a field. Race 10 is uncommon. Surveys in the U.S.A. have not reported this race. Given the variability of race tests (7) and the FI = 9.8 on 'Peking', an FI \geq 10 might be obtained in subsequent tests, thus designating race 14 for the population.

The program at the National Soybean Research Center in Londrina, Paraná, to incorporate resistance to *H. glycines* into soybean includes resistance obtained from 'Peking' (R. A. S. Kiihl, personal communication). Populations of races 2, 4, 5, 10, and 14 identified in this study expressed much higher FI's on 'Pickett 71' than on 'Peking'. Resistant lines that have the 'Peking' source of resistance and are being considered for release in Brazil should be evaluated against these populations to determine if they have a sufficient level of resistance. Management of *H. glycines* should include crop rotation and planting of cultivars resistant to the particular populations present in growers' fields.

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