Evaluation of Soybean Germplasm for Resistance to Soybean Rust (*Phakopsora pachyrhizi*) in Nigeria

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ABSTRACT

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Soybean rust, caused by *Phakopsora pachyrhizi*, is one of the most important constraints to soybean production worldwide. The absence of high levels of host resistance to the pathogen has necessitated the continued search and identification of sources of resistance. In one set of experiments, 178 soybean breeding lines from the International Institute of Tropical Agriculture were rated for rust severity in the field in 2002 and 2003 at Ile-Ife, Yandev, and Ibadan, Nigeria. Thirty-six lines with disease severity ≤3 (based on a 0-to-5 scale) were selected for a second round of evaluation in 2004 at Ibadan. In the third round of evaluation under inoculated field conditions, 11 breeding lines with disease severity ≤2 were further evaluated for rust resistance at Ibadan in 2005 and 2006. The breeding lines TGx 1835-10E, TGx 1895-50F, and TGx 1903-3F consistently had the lowest level of disease severity across years and locations. In another set of experiments, 101 accessions from the United States Department of Agriculture–Agricultural Research Service and National Agriculture Research Organization (Uganda) were evaluated in the first round in 2005 under inoculated conditions in the screenhouse; 12 accessions with disease severity ≤20% leaf area infected were selected for evaluation in the second round in 2005 and 2006 under inoculated field conditions at Ibadan. Highly significant differences ($P < 0.0001$) in disease severity were observed among the 101 accessions during this first round of rust evaluation. Significant ($P < 0.0001$) differences in rust severity and sporulation also were observed among the 12 selected accessions. Accessions PI 594538A, PI 417089A, and UG-5 had significantly ($P < 0.05$) lower disease severity than all other selected accessions in both years of evaluation, with rust severities ranging from 0.1 to 2.4%. These results indicate that some of the breeding lines (TGx 1835-10E, TGx 1895-50F, and TGx 1903-3F) and accessions (PI 594538A, PI 417089A, and UG-5) would be useful sources of soybean rust resistance genes for incorporation into high-yielding and adapted cultivars.

Additional keywords: disease resistance, stability analysis

Soybean rust, caused by *Phakopsora pachyrhizi*, is one of the most economically important foliar diseases affecting soybean worldwide (13). The disease originated from Japan (17) and mainly was associated with Asia and Australia (5). Within the last 10 years, soybean rust was reported in South America (37) and in the continental United States (29). In west and central Africa, soybean rust has been reported in Nigeria, Ghana, and Democratic Republic of Congo (2,3,25). The disease is now endemic in most soybean-producing areas in Nigeria (1,4), the largest producer of soybean in Africa (28). Significant yield losses due to the disease have been reported in Asia (14), South America (37), and Africa, where yield losses of up to 60 to 80% have been reported in the eastern and southern parts of the continent (16,18).

Resistant cultivars and fungicides are the main strategies for managing soybean rust worldwide. Chemical control in commercial soybean plantings adds significantly to production costs (21) and is not a viable option in subsistence production systems in most developing countries in Africa. Deployment of resistant cultivars still remains the most sustainable strategy to manage soybean rust. Specific resistance to *P. pachyrhizi* has been reported, and four single dominant genes have been identified as Rpp_1, Rpp_2, Rpp_3, and Rpp_4 (6,15). These four genes condition resistance to only a limited set of rust isolates. Soybean leaves express three types of reactions depending on a specific pathogen isolate and host resistance interaction. When inoculated with specific isolates, plants with an immune reaction do not produce any visible symptoms. Other resistant plants produce a red-brown (RB) lesion with either no uredinia or only sparsely sporulating uredinia, whereas other genotypes express the susceptible TAN reaction, characterized by tan-colored lesions with many uredinia and abundant sporulation (5). Single-gene resistance may not be durable and its usefulness often is ineffective when exposed to different rust isolates (13).

Renewed efforts to identify resistant genotypes have been initiated in the last 10 years, primarily due to discovery of the pathogen in Africa, South America, and the United States, although screening of soybean germplasm for rust resistance has been ongoing in Asia since 1961. One of the major germplasm screening efforts undertaken recently was by Miles et al. (22), in which over 16,000 soybean accessions in the United States Department of Agriculture–Agricultural Research Service (USDA-ARS) Germplasm Collection were screened for rust resistance. Accessions that had low levels of rust severity have been distributed to breeding programs in Africa and Paraguay to evaluate them against local pathogen populations (4,23). Since early 1970s, many improved soybean lines were developed by the International Institute of Tropical Agriculture (IITA) soybean breeding program that contributed to significant increase in soybean productivity and farm income in Nigeria (24). This program has developed elite breeding lines that are high yielding, possess good agronomic characteristics, and are suitable for commercial production in many parts of Africa (28); however, they have not been systematically evaluated for rust resistance.

Plant breeders routinely test genotypes in multiple locations and years to determine whether or not environment affects the magnitude of specific traits of the genotypes, such as disease severity, as well

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as differences of the values of the traits among genotypes (26). Several methods have been proposed to analyze the genotype–environment (GE) interaction (9,10). Often, a large number of genotypes are tested across a number of sites and years, and it is frequently difficult to determine the pattern of genotypic response across environments without the help of graphical display of the data (34). The biplot technique provides a powerful solution to this problem (11). Biplot analysis is a multivariate analytical technique that graphically displays two-way data and allows visualization of the interrelationship among environments, genotypes, and interactions between genotypes and environments. The genotype main effect plus genotype–environment interaction (GGE) biplot developed by Yan et al. (35) is one of the two types of biplots that has been used to visualize GE two-way data.

This study was conducted to evaluate IITA breeding lines for resistance to *P. pachyrhizi* across different environments, and identify accessions from the USDA-ARS and National Agriculture Research Organization (NARO, Uganda) that are potential sources of rust resistance in Africa.

MATERIALS AND METHODS

Plant material. Two sets of soybean genotypes were evaluated for resistance to soybean rust. The first set consisted of breeding lines from the IITA soybean breeding program, whereas the second set consisted of soybean accessions obtained from the USDA Soybean Germplasm Collection, Urbana, IL and NARO, Uganda. A summary of the pedigrees for the breeding lines and agronomic characteristics for the selected genotypes that were used in this study is presented in Table 1.

Field trials. Soybean seed were planted manually in field plots consisting of two 2-m-rows spaced 0.75 m between rows and 0.2 m within rows. Field trials in 2002, 2003, and 2004 were evaluated under natural *P. pachyrhizi* inoculum pressure. To ensure high and uniform disease pressure in the field in 2005 and 2006, infector rows consisting of a highly susceptible line (TGx 1485-1D) were planted 2 weeks before planting the test lines and placed after every three rows of each test line following a previously reported protocol (31). Plants in the infector rows and the test lines were inoculated twice when test lines were at growth stages V4 and V6. Inoculum was freshly obtained from the spore bank located at IITA field site at Ibadan and prepared as described previously (31). A spore concentration of 1×10^6 urediniospores per milliliter of deionized water containing 0.1% Tween 20 was applied to the abaxial side of the leaves using a 20-liter knapsack sprayer (Cooper Pegler, Burgess Hill, Sussex, UK).

Evaluation of breeding lines. In all, 178 soybean breeding lines were screened for rust resistance in the field in Nigeria at the National Cereal Research Institute substation at Yandev in Benue State and at Ile-Ife in Obafemi Awolowo University Farm in Osun State, in 2002, and at the IITA field site at Ibadan in 2003. The Ile-Ife, Yandev, and Ibadan sites are located in the Derived Savannah agroecological zone, which lies within latitudes 6° 8′ and 9° 30′ N and longitudes 2° 40′ and 12° 15′ E. Plots were planted on 2 and 14 August 2002 at Yandev and Ile-Ife, respectively, and 13 August 2003 at the IITA field site at Ibadan. Genotypes were arranged in a randomized complete block design with two replications. Breeding lines were scored for rust severity when plants were at growth stage R6 and disease severity data were recorded on five randomly selected plants (a composite score considering rust severity of all leaves for each plant) within a row using a 0-to-5 scale (5). From the initial evaluation of 178 breeding lines at the three locations during 2002 to 2003, 36 lines with disease severity of ≤3.0 in all three evaluations were evaluated at Ibadan in 2004.

The selected 36 breeding lines were planted on 13 August 2004 in a randomized complete block design with three replications in the field. Rust was assessed when plants were at growth stage R6. Disease severity data were recorded on five randomly selected plants within a row using a 0-to-5 scale (5). The extent of rust damage on all leaves was considered for recording composite rust score of each plant. Eleven breeding lines that had a disease severity ≤2 in 2004 were evaluated at Ibadan in 2005 and 2006 in the field and

Table 1. Pedigree and agronomic characteristics of selected elite soybean breeding lines from IITA and accessions from USDA-ARS and NARO evaluated for resistance to *Phakopsora pachyrhizi* in Nigeria[a]

Genotype	Type	Origin	Maturity[b]	Days to flowering[c]	Pedigree
TGx 1835-10E	Breeding line	IITA	Early	41	TGx 1213-1D × TGx 1445-3E
TGx 1871-12E	Breeding line	IITA	Early	44	TGx 1740-6F × TGx 1660-15F
TGx 1740-2F	Breeding line	IITA/United States	Early	41	TGx 539-5E × Sibley
TGx 1895-50F	Breeding line	IITA	Medium	43	TGx 1814-3E × TGx 1740-6F
TGx 1891-3F	Breeding line	IITA/Brazil	Medium	44	TGx 1660-19F × BR 839240
TGx 1897-17F	Breeding line	IITA	Early	42	TGx 1809-12E × TGx 1740-6F
TGx 1903-3F	Breeding line	IITA	Early	44	TGx 1740-2F × TGx 1830-32E
TGx 1864-17F	Breeding line	IITA	Late	48	TGx 1447-1D × TGx 1449-2D
TGx 1895-49F	Breeding line	IITA	Early	46	TGx 1814-3E × TGx 1740-6F
TGx 1895-6F	Breeding line	IITA	Early	47	TGx 1814-3E × TGx 1740-6F
TGx 1869-13E	Breeding line	IITA	Late	50	TGx 1448-2E × TGx 1660-15F
TGx 1485-1D[d]	Breeding line	IITA	Early	39	TGx 316-024D × TGx 813-11D
G 00057	Accession	Uganda	Medium	31	…
PI 417089A	Accession	Japan	Medium	33	…
UG-5	Accession	Uganda	Early	42	…
PI 423972	Accession	Japan	Medium	35	…
PI 594538A	Accession	China	Early	31	…
PI 628932	Accession	Brazil	Medium	42	…
G 00072	Accession	Uganda	Early	38	…
PI 368039	Accession	Taiwan	Early	33	…
PI 230970	Accession	Japan	Medium	38	…
PI 594172A	Accession	Japan	Medium	39	…
PI 429329	Accession	Nigeria	Late	44	…
PI 462312	Accession	India	Medium	32	…

[a] Genotypes with TGx, PI, and G or UG prefixes were from the International Institute of Tropical Agriculture (IITA), the United States Department of Agriculture–Agricultural Research Service (USDA-ARS), Soybean Germplasm Collection, Urbana, IL, and the National Agriculture Research Organization (NARO), respectively.

[b] Maturity levels: early = <90 days to maturity, medium = 91 to 100 days to maturity, and late = >100 days to maturity.

[c] Data recorded in 2006 in the field at IITA farm, Ibadan, Nigeria.

[d] TGx 1485-1D is a susceptible breeding line.

screenhouse (only in 2005) under conditions of artificial inoculation with *P. pachyrhizi* following previously reported protocols (31).

The 11 elite breeding lines were planted at Ibadan in field plots on 22 September 2005 and 21 September 2006 and in the screenhouse on 22 July 2005. In the field, lines were arranged in a randomized complete block design with three replications and inoculated as per the infector row system (31). In the screenhouse, seed were planted in 1-m-long rows spaced 0.5 m apart and lines were arranged in a randomized complete block design with four replications. Seedlings were spray inoculated 15 days after emergence with spores collected from a spore bank in the field site. Temperature ranged between 22 and 28°C in the screenhouse, which had no artificial lighting.

Disease severity (percentage of leaf area infected) and sporulation were recorded when breeding lines were at R6 growth stage in the field and the screenhouse. Three individual leaflets at the bottom, middle, and upper layers of five randomly selected plants in each plot were rated individually. Disease severity of the entire plant was based on the mean severity of the three canopy levels. Mean disease severities of five plants were used for data analysis. A sample size of 50 lesions was used to determine the percent sporulation by counting the number of lesions with pustules and expressing this as the percentage of the total sample size. Additional data were recorded on the lesion type (TAN, RB, or a mixture of RB and TAN).

Evaluation of USDA-ARS and NARO germplasm accessions. In total, 101 accessions from USDA-ARS and NARO and susceptible check TGx 1485-1D were evaluated in the screenhouse under inoculated conditions in 2005. Seed were sown in plastic pots on 5 June 2005 and inoculated 15 days after emergence as described earlier (31). Accessions were arranged in a randomized complete block design with two replications. At growth stage R6, data on disease severity (percentage of leaf area infected), percentage sporulation, and lesion type were recorded as described above. From the initial 101 accessions, only 12 with disease severity levels ≤20% leaf area infected were selected for reevaluation under field conditions in 2005 and 2006 at the Ibadan field site.

The 12 selected accessions were planted in the field on 21 and 22 September in 2005 and 2006, respectively, for rust evaluation using the infector system (31). Data on disease severity, sporulation, and lesion type were recorded at growth stage R6 as previously described.

Statistical analysis. Evaluation of breeding lines for rust resistance was conducted in 2002 in Yandev and Ile-Ife and in 2003 to 2006 at Ibadan. The statistical analysis for the breeding lines included an analysis of variance for each environment and a combined analysis across environments using PROC GLM in SAS. In this case, each year–location combination was considered as a unique and random environment, whereas genotype was analyzed as fixed effect. The significance of F ratios was tested according to the procedure described by McIntosh (20) for analysis of combined experiments. Treatment means were separated using Fischer's protected least significance test (LSD).

GGE biplot analyses for rust severity were conducted using GGE biplot software (36) to determine stability and to identify breeding lines of interest for disease resistance. Three environments (Yandev 2002, Ile-Ife 2002, and Ibadan 2003–06) and the 11 elite breeding lines tested across these environments were used in this analysis, with the breeding line TGx 1485-1D as the susceptible check. Disease severity data collected at Ile-Ife, Yandev, and Ibadan using a 0-to-5 scale were converted to percentages using the midpoint method (7) prior to GGE biplot analysis. Field data collected at Ibadan between 2003 and 2006 were averaged across years to provide a single location. A specific option in GGE biplot analysis allows comparison among a set of genotypes with a reference genotype. This method defines the position of an "ideal" genotype, which will have the highest average value of all genotypes and be absolutely stable; that is, it expresses the least GE interaction. A set of concentric circles is generated using the ideal genotype as the concentric center. The ideal genotype is used as a reference to rank the other genotypes. A performance line passing through the origin of the biplot is used to determine mean performance of a genotype. The arrow on the performance line represents increasing mean disease severity (i.e., increase susceptibility to rust). A stability line perpendicular to the performance line also passes through the origin of the biplot; the two arrows in opposite directions represent a decrease in stability. A genotype distanced farther from the biplot origin on either side on the stability line represents relatively lower stability. A genotype closer to the performance line is considered more stable than the one placed farther.

The reaction of USDA-ARS and NARO accessions to soybean rust was evaluated only at Ibadan between 2005 and 2006, and disease severity for the selected accessions was subjected to analysis of variance for each year and combined analysis across years using PROC GLM in SAS. All statistical analyses using SAS were performed in version 9.1 (SAS Institute Inc., Cary, NC).

RESULTS

Evaluation of breeding lines. In 2002, all 178 breeding lines showed rust symptoms and disease severity ranged between 1.0 to 5.0 at Ile-Ife and Yandev, with mean disease severities of 3.2 and 2.1, respectively. Mean disease severity for the same set of soybean lines evaluated at Ibadan in 2003 was 4.6. There were significant differences ($P < 0.0001$) in disease severity among the 178 breeding lines at Ile-Ife and Ibadan but not at Yandev. About 12.4% of all breeding lines had severity ≤2.5 whereas the majority of the lines (62%) had rust severity levels between 3.1 and 4.0. In all, 36 breeding lines with disease severity of ≤3.0 were selected from three sites and reevaluated at Ibadan in 2004. Significant differences ($P < 0.0001$) in disease severity were observed in this first set of selection, with a mean disease severity of 29% leaf area infected. From these 36 lines, 11 elite breeding lines with disease severity level ≤2 were selected for further evaluation at Ibadan between 2005 and 2006.

Within respective years, disease severity differed significantly ($P < 0.0001$) among the 11 elite breeding lines at Ibadan, where the disease pressure was high, but not at Ile-Ife or Yandev (Table 2). The breeding line TGx 1835-10E consistently had the lowest level of disease severity across years and locations, while the susceptible check line TGx 1485-1D had the highest level of disease severity. Other breeding lines that had lower levels of disease severity compared with TGx 1485-1D were TGx 1895-50F and TGx 1903-3F in 2005 and 2006 (Table 2). Levels of sporulation on the breeding line TGx 1835-10E, which had an RB reaction, ranged between 40 and 72% and were significantly ($P < 0.0001$) lower than that on all the other elite breeding lines that had 100% sporulation with TAN reactions (Table 2). Similarly, breeding lines with a TAN reaction (excluding the check line) had significantly ($P < 0.001$) higher levels of disease severity, with a mean of 15% across years compared with TGx 1835-10E, which had an RB reaction and a mean of 6%. Breeding lines TGx 1485-1D flowered earliest (39 days to flowering) whereas TGx 1869-13E had the highest number of days to flowering (50 days). The rest of the breeding lines had intermediate number of days to flowering (Table 1).

Based on the GGE biplot analysis, no significant ($P > 0.05$) GE interaction was detected when the 11 elite breeding lines, including the check line, were evaluated for rust across the three environments. The GGE biplot analysis of the 11 elite breeding and the check line revealed that TGx 1835-10E had the lowest level of rust severity across all locations by being farthest to the left of the biplot origin compared with all the other breeding lines (Fig. 1). In addition, the breeding lines TGx 1895-50F, TGx 1891-3F, and TGx 1903-3F were closer to the point of ideal genotype as well as being higher on mean performance line (Fig. 1). However, TGx 1895-50F and

TGx 1903-3F were much farther to the left side of the origin of the biplot, indicating that they had a much lower level of disease severity compared with TGx 1891-3F. One breeding line, TGx 1869-13E, and the check breeding line, TGx 1485-1D, were consistently the more susceptible by being farthest on the right side of the origin of the biplot on the performance line. Based on the GGE biplot analysis, reaction of breeding lines to rust was more consistent at Yandev but the disease pressure was highest at Ibadan.

Evaluation of USDA-ARS and NARO accessions. Severity of soybean rust for the 101 accessions evaluated in the screenhouse at Ibadan in 2005 differed significantly ($P < 0.0001$) and ranged from 0.1 to 79.7% leaf area infected during this preliminary stage of rust evaluation. Unlike the breeding lines, lesion type on the accessions ranged from hypersensitive to a mixture of RB and TAN reactions. Of the 101 accessions evaluated, 75.3% had a TAN reaction while 14.4, 9.3, and 1% had an RB, RB&TAN, and hypersensitive response (HR) reactions, respectively. A higher level of disease severity was associated with a higher frequency of genotypes with TAN reaction. For example, for disease severity >45% leaf area infected, 64% of the genotypes had a TAN reaction whereas 11.3% had a mixed reaction of TAN and RB. Disease severity <15% was associated only with HR and RB lesion types, with 1 and 4.1% of the total accessions, respectively. Twelve accessions (approximately 12%) had disease severity ≤20% leaf area infected, and these accessions with low rust severity were selected for further evaluation under field conditions at Ibadan between 2005 and 2006.

A significant ($P < 0.0001$) difference in rust severity and sporulation was observed among the selected 12 accessions within each year of evaluation (Table 3). Accessions PI 594538A, PI 417089A, and UG-5 had significantly lower disease severity than all other selected accessions in both years of evaluation, with rust severities ranging from 0.1 to 2.4%. Other accessions with lower levels of disease severity compared with the check breeding line TGx 1485-1D were PI 423972, G 00057, and PI 594172A, with disease severity levels ranging from 3.9 to 9.7%. The corresponding mean rust severity for TGx 1485-1D was 62% leaf area infected. PI 628932 and PI 429329 were the most susceptible of the selected accessions, with disease severities ranging from 13.8 to 15.5% (Table 3). No sporulation was ob-

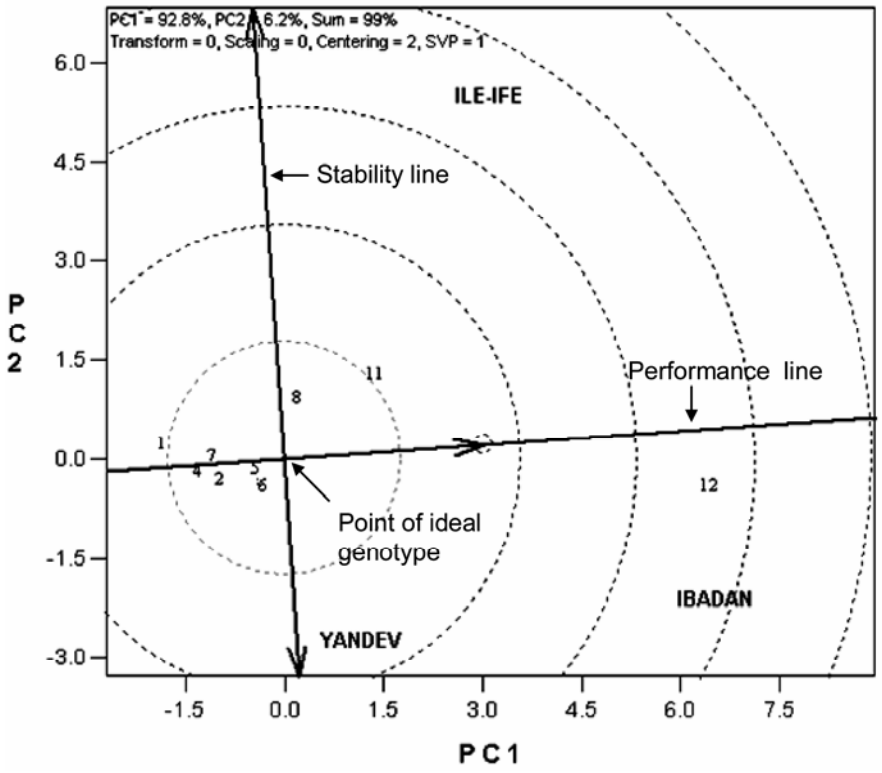

Fig. 1. Genotype and genotype–environment biplot showing a comparison of 12 International Institute of Tropical Agriculture soybean breeding lines with ideal genotypes for rust severity across three environments in Nigeria. The environments are shown in bold and three genotypes (overlapping with those shown in the innermost circle) are hidden for clarity of the graph. (Refer to Table 2 for names of the breeding lines.)

Table 2. Disease severity and lesion infection type on selected breeding lines from a multiyear evaluation of IITA soybean breeding lines for resistance to rust (*Phakopsora pachyrhizi*) in Nigeria[a]

		2002		2003	2004	2005				2006		
		Ile-Ife	Yandev	Ibadan	Ibadan	Ibadan (SH)		Ibadan		Ibadan		
No.	Breeding line	RS	RS	RS	LAI (%)	LAI (%)	Spor (%)	LAI (%)	Spor (%)	LAI (%)	Spor (%)	Type[b]
1	TGx 1835-10E	1.0	2.0	2.0	1.7	9.3	40	4.9	72	7.9	66	RB
2	TGx 1871-12E	1.0	2.5	5.0	1.7	39.5	100	6.0	100	4.3	100	TAN
3	TGx 1740-2F	2.0	2.5	4.5	4.3	36.3	100	14.2	100	8.2	100	TAN
4	TGx 1895-50F	1.0	2.5	3.0	2.3	35.0	100	8.5	100	9.6	100	TAN
5	TGx 1891-3F	2.0	2.0	4.0	3.3	14.6	100	10.3	100	11.5	100	TAN
6	TGx 1897-17F	1.5	2.5	5.0	2.7	32.3	100	14.5	100	11.5	100	TAN
7	TGx 1903-3F	2.0	2.5	…	1.0	15.3	100	11.6	100	13.7	100	TAN
8	TGx 1864-17F	3.0	2.0	3.5	8.7	14.8	100	15.0	100	15.0	100	TAN
9	TGx 1895-49F	1.5	2.5	5.0	5.0	22.9	100	8.9	100	15.1	100	TAN
10	TGx 1895-6F	1.0	2.0	…	9.7	32.5	100	20.3	100	16.2	100	TAN
11	TGx 1869-13E	3.0	2.0	4.0	11.3	34.6	100	23.8	100	19.0	100	TAN
12	TGx 1485-1D[c]	2.0	3.0	5.0	58.3	64.3	100	65.3	100	64.1	100	TAN
…	LSD ($\alpha = 0.05$)[d]	2.4	0.9	1.1	9.2	5.3	3.0	2.6	9.0	1.8	1.5	…

[a] In all years, evaluations were all conducted under field conditions except in 2005, where evaluations were conducted under screenhouse (SH) and field conditions. RS denotes rust severity based on a rating scale of 0 to 5, LAI denotes leaf area infected, and Spor denotes sporulation, which is based on a sample of 50 lesions.

[b] Lesion type: RB denotes reddish-brown hypersensitive lesions with 0 to 2 pustules per lesion and TAN denotes tan-colored lesions with 2 to 6 pustules per lesion.

[c] TGx 1485-1D was used as the susceptible check line in all locations.

[d] LSD = least significant difference.

served on PI 594538A, which had an HR reaction, whereas ≤10% sporulation was observed on PI 417089A and UG-5, both with an RB reaction. All the remaining accessions had significantly ($P < 0.05$) higher levels of sporulation with RB reactions, except PI 429329 and PI 230970, which had a mixed reaction of RB and TAN (Table 3). PI 594538A and G 00057 were earliest to flower whereas PI 429329 had the highest number of days to flowering, with the remaining accessions having intermediate number of days to flowering (Table 1).

DISCUSSION

Deployment of resistant cultivars still remains the most viable strategy to manage soybean rust in Africa and other countries in the developing world. In this study, soybean breeding lines developed by IITA and germplasm developed elsewhere with previously unknown resistance under our conditions were evaluated in different locations to identify genotypes that are resistant to soybean rust across geographical areas in Nigeria. Breeding lines and accessions from USDA-ARS and NARO with low levels of rust resistance were identified across years and locations. This is the first study in West Africa to document soybean genotypes with high levels of resistance to *P. pachyrhizi*. Such breeding lines with low rust severity may be used directly for commercial cultivation while accessions with low disease severity could be useful sources of resistance genes for soybean breeding programs. Mean disease severity differed across years for the selected elite breeding lines and accessions, and this may be due to differences in environmental conditions across time and sites that influence disease development (27). Although all the sites are located in the Derived Savannah agroecological zone, they differ greatly in key attributes such as temperature and rainfall that could affect disease expression (8).

Among the soybean breeding lines, TGx 1835-10E had the lowest level of rust severity across the three environments. In addition, this breeding line also had the lowest levels of sporulation and was the only line that had an RB reaction. TGx 1835-10E has been released for general cultivation in Uganda as rust-resistant cv. MAKSOY 1N (P. Tukamuhabwa, *personal communication*). Similarly, the accessions PI 417089A and UG-5, which had very low levels of disease and sporulation, had an RB reaction type. Soybean genotypes expressing the RB reaction have been associated with single-gene resistance (12). In previous studies, host–pathogen combinations that resulted in RB reactions tended to have longer latent periods, lower rates of increase in pustule number over time, and smaller lesions compared with susceptible interactions that resulted in a TAN reaction (5,19), and these parameters are characteristic of partial resistance (33). Thus, these genotypes with low rust severities with RB reactions may be sources of partial resistance to *P. pachyrhizi*. It has been observed that identification and utilization of partial resistance in breeding program has been limited (22) and more research is needed to fully utilize partial resistance traits. Accessions PI 594538A and PI 417089A previously were reported as resistant to rust in the United States (22) whereas UG-5 has been reported to have high levels of rust resistance in Uganda (16). Although previous evaluations of PI 594538A reported a TAN reaction type (21), we observed only minute flecks similar to an HR reaction. In addition, PI 417089A was reported to have a TAN reaction (32) whereas an RB reaction was observed in this study. These differences may be attributed to pathogenic variation in rust isolates (12) that were used for inoculation. Miles et al. (22) used a mixture of four isolates from Thailand, Brazil, Paraguay, and Zimbabwe, while Voung et al. (32) used nonpurified rust isolates from soybean in Vietnam. In the present study, we used natural inoculum in the field experiments, while *P. pachyrhizi* inoculum prepared from infected soybean leaves collected from the IITA research farm was used for artificial inoculations in the screenhouse experiments.

Analysis of the stability of soybean breeding lines for rust resistance using GGE biplot technique showed that eight breeding lines in the innermost circle could be considered stable for rust resistance across the three environments. All these breeding lines could be of value for breeding programs attempting to improve soybean rust resistance. TGx 1895-50F and TGx 1903-3F had the lowest levels of disease severity and would be the preferred genotypes for the breeding programs. Although the stability analysis was not performed on the USDA-ARS and NARO accessions due to the single location of disease evaluation, the very low levels of disease severity observed for accessions PI 594538A, PI 417089A, and UG-5 make these additional sources of resistance to be considered in a rust-resistance breeding program.

Identification of genotypes that possess high stability for low disease severity is a key component that ensures the selection of useful sources of high resistance for breeding programs (30). A regular stability analysis often does not provide relative ranking of superior entries in reference to an ideal genotype, which results in a sub-

Table 3. Performance of selected germplasm during evaluation of soybean accessions from USDA-ARS and NARO for resistance to soybean rust, *Phakopsora pachyrhizi*, at Ibadan, Nigeria[a]

	2005				2006		
	Screenhouse		Field		Field		
Soybean accession	LAI (%)	Spor (%)	LAI (%)	Spor (%)	LAI (%)	Spor (%)	Lesion type[b]
PI 594538A	0.1	0	0.1	0	0.1	0	HR
PI 417089A	5.3	10	0.2	4	0.7	6	RB
UG-5	6.0	20	2.0	6	2.4	10	RB
PI 594172A	17.2	20	6.8	23	9.7	64	RB
PI 423972	22.5	40	3.9	29	7.7	64	RB
G 00057	16.3	40	7.9	21	5.4	28	RB
PI 462312	22.2	40	11.8	72	9.1	66	RB
PI 368039	25.8	90	8.1	53	7.0	33	RB
PI 628932	19.6	60	14.9	61	15.5	38	RB
G 00072	33.7	85	8.3	60	14.0	80	RB
PI 429329	23.6	80	13.8	78	14.5	89	RB & TAN
PI 230970	35.0	84	9.5	61	6.5	72	RB & TAN
TGx 1485-1D[c]	64.3	100	60.0	100	64.1	100	TAN
LSD ($\alpha = 0.05$)[d]	5.6	6.5	2.0	9.0	1.8	9.2	…

[a] USDA-ARS = United States Department of Agriculture–Agricultural Research Service, Soybean Germplasm Collection, Urbana, IL; and NARO = National Agriculture Research Organization, Uganda. LAI denotes leaf area infected and Spor denotes sporulation, which is based on a sample 50 lesions.

[b] HR = hypersensitive response, with small, hypersensitive, pin-head like specks; RB represents reddish-brown lesions, with 0 to 2 pustules per lesion; and TAN indicates tan-colored lesions with 2 to 6 pustules per lesion.

[c] TGx 1485-1D was used as the susceptible check.

[d] LSD = least significant difference.

jective judgment when selecting a cultivar (35). The GGE biplot approach used in this study could help breeders better prioritize genotypes to use. The combined visual assessment of the level of resistance and its stability is a big advantage, and adds confidence in the decision to promote a superior genotype. This GGE biplot approach has been used in selection of superior wheat genotypes that have low and stable resistance to spot blotch (30).

This study identified specific breeding lines from the IITA breeding program and accessions from USDA-ARS and NARO with very high levels of resistance to soybean rust. The absence of high levels of genetic resistance to the pathogen dictates the continued search and identification of sources of resistance genes. The soybean breeding lines TGx 1835-10E, TGx 1895-50F, and TGx 1903-3F can be used directly as commercial cultivars in West Africa due to superior yield performance (*data not presented*). Accessions PI 594538A, PI 417089A, and UG-5 can be used as sources of resistance genes in soybean breeding programs for the development of genotypes with good levels of resistance to *P. pachyrhizi* which are adapted to West African conditions. Additionally, crosses between PI 594538A, PI 417089A, and UG-5 and the susceptible breeding line TGx 1485-1D have generated several progenies with high levels of rust resistance, and preliminary data are reported elsewhere (1,4).

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