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Molecular marker-based genetic diversity assessment of *Striga*-resistant maize inbred lines

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Abstract *Striga*-resistant maize inbred lines are of interest to maize breeding programs in the savannas of Africa where the parasitic weed is endemic and causes severe yield losses in tropical maize. Assessment of the genetic diversity of such inbred lines is useful for their systematic and efficient use in a breeding program. Diversity analysis of 41 *Striga*-resistant maize inbred lines was conducted using amplified fragment length polymorphism (AFLP) and simple sequence repeat (SSR) markers to examine the genetic relationships among these lines and to determine the level of genetic diversity that exists within and between their source populations. The two marker systems generated 262 and 101 polymorphic fragments, respectively. Genetic similarity (GS) values among all possible pairs of inbred lines varied from 0.45 to 0.95, with a mean of 0.61 ± 0.002 for AFLPs, and from 0.21 to 0.92, with a mean of 0.48 ± 0.003, for SSRs. The inbred lines from each source population exhibited a broad range of GS values with the two types of markers. Both AFLPs and SSRs revealed similar levels of within population genetic variation for all source populations. Cluster and principal component analysis of GS estimates with the two markers revealed clear differentiation of the *Striga*-resistant inbred lines into groups according to their source populations. There was clear separation between early- and late-maturing *Striga*-resistant inbred lines. Considering the paucity of germplasm with good levels of resistance to *Striga* in maize, the broad genetic diversity detected within and among source populations demonstrates the genetic potential that exists to improve maize for resistance to *Striga*.

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Introduction

Striga is a parasitic weed posing a serious threat to cereal production in sub-Saharan Africa. The problem of *Striga* is intensifying across regions in sub-Saharan Africa because of deteriorating soil fertility, shortening of the fallow period, expansion of production into marginal lands with little nutrient input and an increasing trend towards a continuous cultivation of one crop in place of traditional rotation and inter-cropping systems (Kling et al. 2000). *Striga* severely affects an estimated 40 million hectares of land devoted to cereal production in West Africa alone, with an additional 70 million hectares having moderate levels of infestation (Lagoke et al. 1991). Heavy infestation with *Striga* can render land unfit for crop production, and fields have been abandoned in the worst affected areas. The effects of this parasite are likely to be long lasting as *Striga* plants produce millions of tiny seeds that can stay viable in the soil for many years.

Among the numerous species of *Striga* that are endemic to Africa, *Striga hermonthica* (Del.) Benth is the most widespread species attacking cereals and causes the greatest economic damage (Ramaiah 1987; Efron et al. 1989; Lagoke et al. 1991; Berner et al. 1995). Maize is a relatively new crop in the African savannas where *Striga* is endemic, and most tropical maize varieties are not expected to be resistant to *Striga* in this region (Ramaiah 1987; Ejeta and Butler 1993). As the maize cultivars currently grown by farmers may suffer a 100% yield loss under heavy infestation (Kling et al. 2000), resistance to *Striga* is an important trait for maize cultivars specifically developed for the savannas. Host plant resistance is regarded as the most economically feasible and practical

means to reduce losses to *Striga* and should be central to any integrated *Striga* control approach recommended to farmers. The advances made in the development of effective, efficient and reliable artificial field infestation techniques at the International Institute of Tropical Agriculture (IITA) in the late 1980s facilitated the identification of inbred lines and hybrids with some level of polygenetic resistance to *S. hermonthica* from diverse sources of germplasm (Kim 1991; Kim and Winslow 1991). These inbred lines were then used as sources of *Striga* resistance to develop composites, synthetics and a backcross population, which have subsequently been improved for resistance to *S. hermonthica* through recurrent selection (Kling et al. 2000).

S. hermonthica is an allogamous species with significant genetic variation within and between populations from different regions of Africa (Musselman et al. 1991; Koyama 2000). To ensure that the maize germplasm developed at IITA possesses resistance to potentially different populations of *S. hermonthica*, the screening and testing of genetic materials under artificial *Striga* infestation have been conducted in multiple locations over seasons (Berner et al. 1995). Repeated screening of lines extracted from improved cycles of selection of composites, synthetics and a backcross population in the field and in the screen house under artificial *Striga* infestation has yielded resistant inbred lines (Kling et al. 2000). Since these source populations share some common parentage in their genetic backgrounds, additional information on the genetic relationships among the *Striga*-resistant inbred lines derived from them would be useful in order to improve the ability to successfully breed maize for broad and durable resistance to *S. hermonthica*. The accurate assessment of the genetic structure and the level of genetic diversity of these inbred lines would be useful for the efficient selection of parental genotypes for crossing (Russell et al. 1997), thereby ensuring long-term and sustained gain from selection for resistance to *Striga* and providing new sources of genetic variation to maize breeders for further genetic advances. Consequently, the utilization of molecular markers that allow direct assessment of genetic differences between these inbred lines may facilitate the development of a systematic and efficient breeding strategy aimed at improving maize for resistance to *Striga*.

The use of different classes of molecular markers may provide robust estimates of the genetic diversity of tropical germplasm having a mixed genetic composition and a broad genetic base (Pinto et al. 2003). A combination of different kinds of markers can also generate sufficient polymorphism to assess genetic relationships even within closely related species (Guadagnuolo et al. 2001). Simple sequence repeat (SSR) markers have been used for genetic diversity assessment studies in maize due to their ability to provide high levels of polymorphism and reliable and reproducible results (Senior et al. 1998; Bernardo et al. 2000; Enoki et al. 2002; Matsuoka et al. 2002; Warburton et al. 2002; Reif et al. 2003). Amplified fragment length polymorphism (AFLP), a class of markers developed by Vos et al. (1995), has also been used to assess the genetic relationships among maize inbred lines (Ajmone-Marsan et al. 1998; Pejic et al. 1998; Lôbberstedt et al. 2000; Barbosa et al. 2003). The diversities of the SSR and AFLP markers have been strongly correlated with restriction fragment length polymorphism (RFLP) diversity and show good agreement with pedigree data in temperate maize (Smith et al. 1997; Lôbberstedt et al. 2000). A comparative study of AFLP and SSR markers in maize generally showed good agreement between the genetic patterns revealed by the two molecular marker types (Pejic et al. 1998).

The objectives of the study reported here were: (1) to examine the genetic relationships among 41 *Striga*-resistant inbred lines derived from four source populations based on AFLP and SSR markers and (2) to determine the level of genetic diversity which exists within and between these source populations.

Materials and methods

Genetic materials

A total of 41 *Striga*-resistant inbred lines at the S_6 to S_9 generations developed at IITA were used. As shown in Table 1, the number of inbred lines extracted from the TZL COMP1, TZE COMP5, STR SYN-W/Y and *Zea diploperennis* (*Z. diplo*) BC_4 germplasms were 8, 12, 10, and 11, respectively. The genetic background of each source population summarized in Table 1 has been fully described by Kling et al. (2000) and Kim et al. (1998). TZL COMP1 and TZE COMP5 share four inbred lines used as *Striga*-resistant donor parents. STR SYN-Y/W and TZL COMP 1 were used as two of the four recurrent parents during the development of the *Z. diplo* BC_4 population..Repeated screening of selected lines from each of the four source populations under artificial infestation with *Striga hermonthica* both in the screen house and in the field resulted in the identification of the 41 *Striga*-resistant inbred lines used in this study.

Laboratory analyses

Fifteen to twenty 10-day-old seedlings of each inbred line (Table 1) were harvested, frozen, and ground into fine powder. The DNA was extracted using the CTAB procedure (Saghai Maroof et al. 1994). Thirty-one SSR primers (bnlg105, bnlg1014, bnlg1035, bnlg1037, bnlg1057, bnlg1108, bnlg1185, bnlg1360, bnlg1754, bnlg1917, bnlg 2126, dupssr11, dupssr15, dupssr19, dupssr23, macE01F06, mag1F03, mmc0081, mmc0121, nc030, phi034, phi037, phi064, phi080, phi126, umc1029, umc1014, umc1015, umc1041, umc1154 and umc1305) were purchased from Research Genetics (Huntsville, Ala.). PCR analyses were performed in 15-µl reaction

Table 1 List of source populations for the 41 *Striga*-resistant inbred lines used in this study

Source population	Genetic background of source population	Tested lines
TZL COMP1	A late-maturing (120 days) composite formed bycrossing TZB-SR with seven *Striga*-resistant inbred lines.The composite has undergone six cycles of recurrent selection under artificial infestation with *S. hermonthica* at two locations in Nigeria	C101–C108
TZE COMP5	An early-maturing (95 days) composite formed by crossing TZESR-W C3 with ten *Striga*-resistant inbred lines. The composite has undergone seven cycles of recurrent selection under artificial infestation with *S. hermonthica* at two locations in Nigeria	C501–C512
STR SYN-Y/W	Half-sib families derived from two synthetics, STR SYN-W and STR SYN-Y, formed from four white and four yellow *Striga*-resistant inbred lines, respectively, were inter-crossed to form STR SYN-Y/W. This synthetic was improved for resistance to *Striga* under artificial infestation with *S. hermonthica* in the field at two locations in Nigeria	SY01–SY10
Z. diplo BC_4	A *Zea diploperennis* accession that supported little or no *S. hermonthica* emergence was crossed to normal maize adapted to West and Central Africa. The resulting F_1 was backcrossed to four different adapted maize genotypes (9022-13, TZL COMP 1, SUWAN 1-SR, and STR SYN-W) in the screen house at Ibadan to form a *Z. diplo* BC_4 population	ZD01–ZD11

mixtures containing 50 ng genomic DNA template, 10 m*M* Tris-HCl (pH 8.3), 50 m*M* KCl, 2.0 m*M* $MgCl_2$, 0.1% Triton X-100, 0.2 μM each dNTP, 0.2 μM each oligonucleotide primer, and 1 U *Taq* DNA polymerase. The amplification was carried out in a 96-well DNA Thermal Cycler (MJ Research, Watertown, Mass.) using a ''touchdown'' program. The thermocycling profile included an initial denaturation of 94°C for 5 min, followed by (1) denaturing at 94°C for 45 s, (2) annealing at 65°C for 60 s and (3) extension at 72°C for 60 s. The annealing temperature was decreased from 65°C to 55°C with a 10°C drop per cycle. The reactions were subjected to 25 additional cycles after reaching the final annealing temperature. This was followed by a final extension at 72°C for 7 min. Amplified products were mixed with 5 µl of loading dye (60% sucrose, 1 m*M* cresol red dye) and resolved on 2% MetaPhor agarose gels (FMC-Bioproducts, Rockland, Me.). The gels were run at 100 V for 6 h in 1× TBE buffer (89 m*M* Tris-Borate plus 2 m*M* EDTA, pH 8.5) and then stained with 0.1% ethidium bromide for 20 min, visualized over a transilluminator and photographed under UV-light. Only clear polymorphic SSR bands with different sizes were scored manually in a binary form as 1 or 0 for their presence or absence, respectively, in each line.

The AFLP analysis was performed following the standard procedure described by Vos et al. (1995). Genomic DNA (0.3 µg) of each line was digested with a pair of restriction enzymes (*Eco*RI and *Mse*I) and ligated to double-stranded adapters. The ligate was preamplified with nonselective primers, and selective amplification was carried out using pairs of selective primers with *Eco*RI/*Mse*I extensions (E32/M49, E32/M50, E32/M59, E32/M60, E32/M61, E33/M48, E33/M60, E33/M61, E36/M47, E36/M48, E36/M50, E38/M47, E38/M59, E40/M47, E40/M48, E40/M49, E41/M47, E41/M48, E41/M59, E41/M62). The products were separated on polyacrylamide gels and silver stained following the instructions from Promega (Madison, Wis.). Only polymorphic bands of a strong intensity were scored manually in binary form as 1 or 0 for their presence or absence, respectively, in each line.

Statistical analysis

Frequencies of polymorphic fragments detected with 20 AFLP primer combinations and 31 SSR primers were calculated for the four source populations of the *Striga*-resistant inbred lines. Differences in the distribution of genetic diversity present within the source populations were estimated as AFLP and SSR diversity based on Nei's (1987) genetic diversity as: $h = n_I(1-\sum x_I^2)/(n_I-1) = (2pqn)/(n-1)$, where x_I was the frequency of a polymorphic fragment at the Ith locus, p was the frequency of the presence of a fragment and q was the frequency of the absence of fragment. Genetic similarities (GS) between pairs of *Striga*-resistant inbred lines were calculated from the 262 AFLP and 101 SSR fragments using a SAS macro (Mumm and Dudley 1995) based on the formula developed by Dice (1945). The average GS and standard deviations within and between source populations were obtained by the MEANS procedure in SAS (SAS Institute 1997). The AFLP-based GS matrix was compared with the SSR-based GS matrix by the MAXCOMP routine of NTSYS-PC (Rohlf 1998) using Mantel's test (1967). Dendrograms were constructed from the GS matrixes by the UPGMA clustering method with the NTSYS-PC package (Rohlf 1998) to visualize the patterns of diversity among the 41 inbred lines. Bootstrap analysis was performed using the software package WINBOOT developed at IRRI (International Rice Research Institute, Manila, The Philippines) with the number of iterations set at 1,000 (Yap and Nelson 1996). The cophenetic correlation coefficients were calculated, and Mantel's test (Mantel 1967) was performed to check the goodness of fit of each UPGMA clustering to the GS matrix on which it was based using the appropriate routines of NTSYS-PC package (Rohlf 1998). Additionally, principal component analyses were calculated from the GS matrices using PC SAS (SAS Institute 1997).

Results

Extent of polymorphism detected with AFLP and SSR markers

Profile analyses of the 41 *Striga*-resistant inbred lines were carried out with AFLP and SSR markers. The 20 AFLP primer pairs generated 262 distinguishable polymorphic fragments. The number of polymorphic fragments for each AFLP primer pair varied from 8 to 21, with an average of 13 per primer pair. The 31 SSR primers generated a total of 101 polymorphic fragments. The number of alleles per SSR locus ranged from two to eight, with an average of three per locus. The proportion of polymorphic AFLP fragments detected in each source population varied from 79% for STR SYN-W/Y to 84% for *Z. diplo* BC_4. The highest level of polymorphic SSR fragments was found in STR SYN-W/Y (81%), while the lowest level was detected in TZL COMP1 (73%) (Table 2). Less than 30% of the polymorphic fragments detected in single source populations with each marker occurred at low frequencies (<0.25). Over 60% of the AFLP and about 50% of the SSR fragments represented a broad range of frequencies in each source population (Table 2).

Genetic diversity in source populations

Among the polymorphic AFLP fragments that were unique to each source population, four were obtained in TZE COMP5 and two in *Z. diplo* BC_4. These fragments occurred at frequencies varying from 0.33 to 0.83. Furthermore, one AFLP allele was absent in all of the inbred lines derived from TZE COMP5 but was present in all of the inbred lines from the other source populations. Eleven polymorphic SSR fragments were unique to single source populations, of which three were observed in TZL COMP1, four in TZE COMP5, three in STR SYN-W/Y and one in *Z. diplo* BC_4 at low frequencies (<0.40). The frequencies of polymorphic fragments detected with AFLP and SSR markers were used to estimate the level of genetic diversity within each source population (Table 3). The source populations exhibited similar levels of Nei's (1987) genetic diversity among the sampled inbred lines with the two marker types.

Diversity patterns among the *Striga*-resistant inbred lines

The GS estimates for pairs of *Striga*-resistant inbred lines within and between source populations calculated

Table 2 Frequencies of amplified products generated with 20 AFLP primer pairs and 31 SSR primers in different source populations for *Striga*-resistant maize inbred lines

Germplasm	Number of fragments with frequencies:					
	AFLP			SSR		
	0.0 and 1.0	0.01–0.24	0.25–0.92	0.0 and 1.0	0.01–0.24	0.25–0.92
TZL COMP1	52	33	177	26	18	57
TZL COMP5	49	72	164	21	26	54
STR SYN-W/Y	55	40	167	19	28	54
Z. diplo BC_4	41	49	172	27	26	48

Table 3 Genetic diversity estimates among *Striga*-resistant maize inbred lines within source populations determined with AFLP and SSR markers using Nei's (1987) genetic diversity (*SE* standard error)

Source	AFLP		SSR	
	Range	Mean (±SE)	Range	Mean (±SE[a])
Nei's Genetic diversity estimates				
TZL COMP1	0.00–0.57	0.35± 0.01	0.00–0.57	0.35± 0.03
TZL COMP5	0.00–0.55	0.31± 0.01	0.08–0.51	0.32± 0.02
STR SYN-W/Y	0.00–0.56	0.32± 0.01	0.00–0.56	0.36± 0.02
Z. diplo BC_4	0.00–0.55	0.34± 0.01	0.00–0.53	0.29± 0.03

for each marker system are shown in Table 4. The GS values for all possible pairs of lines ranged from 0.45 to 0.95, with a mean of 0.61 ± 0.002 for AFLPs, and from 0.21 to 0.92, with a mean of 0.48 ± 0.003, for SSRs. The average GS for pairs of lines within each source population ranged from 0.64 to 0.70 for AFLPs and from 0.49 to 0.53 for SSRs. Pairs of inbred lines from each source population exhibited a broad range of GS values with each marker system (Table 4). The smallest range of GS values among inbred lines was recorded in STR SYN-W/Y, while the largest ones were recorded in TZL COMP1 and *Z. diplo* BC_4 with the two marker types. The average GS values between pairs of lines within a given source population was higher than the average GS values between pairs of lines from different source populations (Table 4). Pairs of lines from different combinations of source germplasm had average GS values varying from 0.56 to 0.61 for AFLPs and from 0.42 to 0.50 for SSRs.

Cluster analysis of the AFLP-based GS estimates provided a fairly good separation of the *Striga*-resistant inbred lines on the basis of their source populations (Fig. 1). The dendrogram derived from AFLP markers separated the lines into four groups, at about a 58% similarity level. Six of the eight inbred lines included in the first group (G-I) were derived from TZL COMP1, and all of the inbred lines clustered in the second group (G-II) were extracted from TZE COMP5. The third group (G-III) contained nine of the ten inbred lines derived from STR SYN-W/Y. The fourth group (G-IV) represented a tight cluster of six inbred lines from *Z. diplo* BC_4 and an inbred line from STR SYN-W/Y. The two AFLP groups, G-II and G-IV, were supported by high bootstrap values (Fig. 1). Although the correlation between the GS matrixes obtained with the two marker systems was low ($r = 0.47$, $P < 0.001$), the dendrogram constructed from SSR-based GS estimates showed genetic relationships similar to those depicted by the AFLP markers, with some differences in assigning the inbred lines into the main groups (Fig. 2). For example, all of the inbred lines from TZL COMP1 that clustered in G-I with AFLPs were assigned to G-I and G-IV with SSRs. The two marker systems clustered all inbred lines from TZE COMP5 into one group (G-II), except for one line that was assigned to G-IV with SSR. G-II of the SSRs also included one line from TZL COMP1 and two lines from STR SYN-W/Y. The inbred lines from STR SYN-W/Y and *Z. diplo* BC_4 clustered in G-III with SSRs but were separated into two groups with AFLPs. In general, the bootstrap probability values for nodes formed with SSRs were low. The cophenetic correlation coefficient between the dendrogram and the original GS matrix showed a good fit for AFLPs ($r = 0.86$) but not for SSRs ($r = 0.74$).

The genetic relationships among the 41 *Striga*-resistant inbred lines determined using principal component analysis of AFLP-based GS estimates are presented in Fig. 2. The first (PC1) and second (PC2) principal component axes accounted for 27% and 12% of the total variation, respectively. The two axes separated the inbred lines into four distinct groups consistent with the results of cluster analysis (Fig. 2). The two principal

Table 4 Range, mean and standard deviation (SD) of Dice's (1945) genetic similarity (GS) estimates among *Striga*-resistant inbred lines within and between source populations calculated from 262 AFLP and 101 SSR markers

Combination source populations	Number of pairwise comparisons	AFLP		SSR	
		Range	Mean (±SD)	Range	Mean (±SD)
C1C1[a]	28	0.51–0.95	0.64 ± 0.10	0.31–0.92	0.49 ± 0.13
C5C5[b]	66	0.58–0.86	0.70 ± 0.07	0.37–0.85	0.56 ± 0.11
SYSY[c]	45	0.56–0.76	0.68 ± 0.04	0.38–0.68	0.53 ± 0.08
ZDZD[d]	55	0.48–0.90	0.64 ± 0.11	0.40–0.91	0.58 ± 0.13
C1C5	96	0.50–0.68	0.59 ± 0.03	0.21–0.65	0.44 ± 0.08
C1SY	80	0.53–0.68	0.61 ± 0.03	0.26–0.56	0.42 ± 0.07
C1ZD	88	0.47–0.68	0.58 ± 0.05	0.28–0.62	0.46 ± 0.07
C5SY	120	0.49–0.68	0.59 ± 0.03	0.29–0.67	0.47 ± 0.07
C5ZD	132	0.45–0.66	0.56 ± 0.04	0.29–0.62	0.47 ± 0.07
SYZD	110	0.50–0.74	0.61 ± 0.05	0.31–0.68	0.50 ± 0.08

[a] C1, TZL COMP1
[b] C5, TZL COMP5
[c] SY, STR SYN-W/Y
[d] ZD, *Z. diplo* BC_4

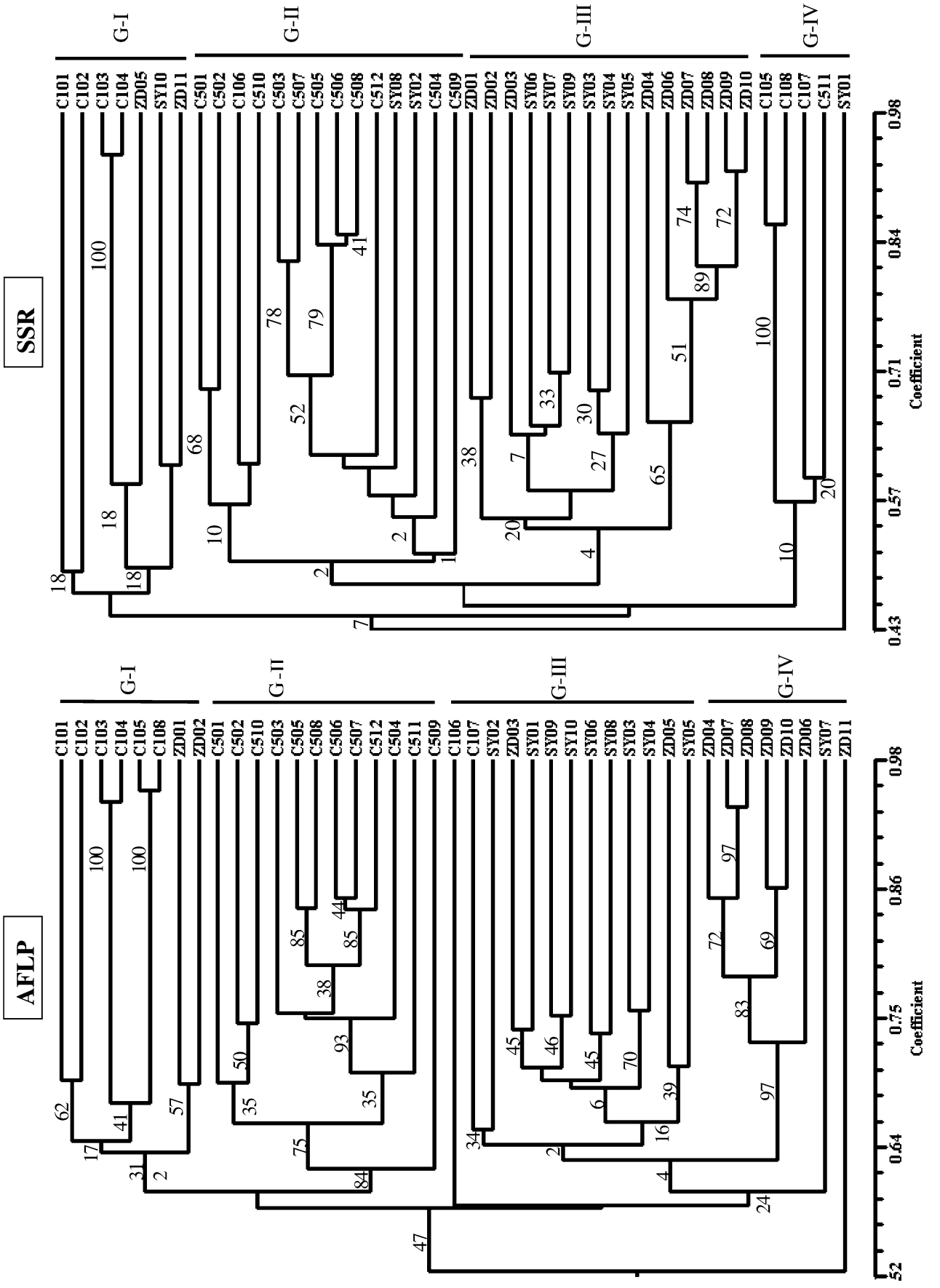

Fig. 1 Dendrogram of 41 *Striga*-resistant inbred lines obtained using AFLP and SSR markers separately

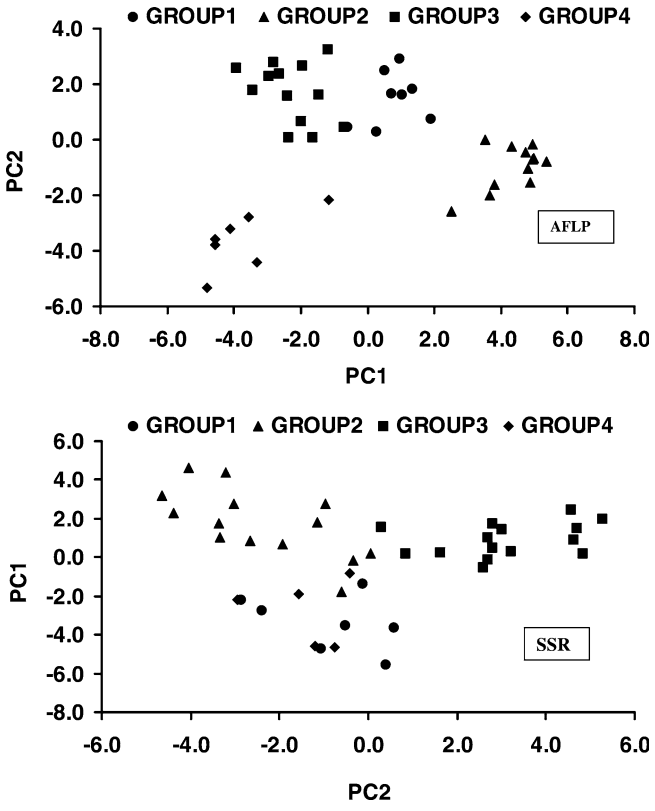

Fig. 2 Scatter plot of *Striga*-resistant maize inbred lines derived from four source populations determined on the basis of principal component analysis of AFLP- and SSR-based estimates of Dice's (1945) genetic similarities (GS)

component axes derived from SSR-based GS estimates also showed a clear separation of G-II (group2) and G-III (group3) (Fig. 2). However, G-I (group1) and G-IV (group4) overlapped and formed one group, which was separated from the other two groups. In the SSR data, the PC1 and PC2 accounted for 19% and 15% of the total variation, respectively (Fig. 2).

Discussion

We employed AFLP and SSR markers to assess the genetic structure and relationships among 41 *Striga*-resistant maize inbred lines. These markers differ in the nature of the evolutionary mechanisms underlying their variation and distribution in the genome (Powell et al. 1996; Staub et al. 1996). In the present study, SSR markers detected higher levels of genetic diversity among the *Striga*-resistant inbred lines within and among source populations than the AFLP markers, which is consistent with the findings from other studies (Powell et al. 1996; Russell et al. 1997; Pejic et al. 1998; Teulat et al. 2000; Barbosa et al. 2003). Such differences between the two marker systems may result from higher levels of mutation in SSR regions than in other parts of the genome (Jarne and Lagoda 1996). The high level of genetic diversity revealed by SSRs may further contribute to the observed low correlation between GS estimates generated with SSR and AFLP markers, since GS estimates are generally lower with SSR than with other markers (Powell et al. 1996; Russell et al. 1997). Bohen et al. (1999) speculated that the low correlation between AFLP- and SSR-based GS estimates may be due to a low linkage between marker loci from different marker systems resulting in sampling of different parts of the genome. In spite of these differences, however, both AFLP and SSR markers revealed similar patterns of the genetic relatedness of the *Striga*-resistant inbred lines in the present study, which is consistent with the findings from other comparative studies (Ajmone-Marson et al. 1998; Mumm and Dudley 1994; Smith et al. 1997; Barbosa et al. 2003).

This study demonstrates that there is considerable diversity among the *Striga*-resistant inbred lines within each source population. Inter-crossing of diverse parental materials during the formation of each source population might have contributed to the observed high level of within-population diversity. Both AFLP and SSR markers revealed similar levels of within-population genetic variation for all source populations, which was unexpected since the source populations were formed from adapted germplasm with different genetic backgrounds notwithstanding the use of some lines as common sources of genes for resistance to *Striga* (Kling et al. 2000). While STR SYN-W/Y is a narrow-based synthetic formed from eight inbred lines, other source populations were formed from adapted germplasm with broad genetic bases. Thus, the level of within-population genetic diversity detected with these markers may not reflect the potential diversity available in each source germplasm because the lines included in the present study were not selected to accurately represent the different source populations. Further diversity analyses using more representative samples of inbred lines from each source population may be needed to accurately estimate the extent of within-population genetic diversity existing in these source populations.

AFLP and SSR markers differentiated the *Striga*-resistant inbred lines into groups representing the different source populations, which is in agreement with data available for maize (Ajmone-Marson et al. 1998; Mumm and Dudley 1994; Smith et al. 1997; Barbosa et al. 2003). There was clear separation between early-maturing inbred lines from TZE COMP5 and late-maturing inbred lines from TZL COMP1, STR SYN-W/Y and *Z. diplo* BC_4. Also, most of the late-maturing inbred lines from the same source population grouped together, with only a few lines clustering with lines from other source populations. Such a separation of the inbred lines on the basis of their source populations could be mainly due to differences in the frequencies of the AFLP and SSR fragments common among the source populations. Significant changes in allele frequencies in different directions could occur

during intensive screening of the source populations for resistance to *Striga* at different cycles of recurrent selection. Labate et al. (1999) indicated that a large fraction of loci in the maize genome could be affected by selection.

Although the overall portrayal of the genetic relationships among the inbred lines tested with the two marker types was in good agreement, AFLP markers provided a better resolution of genetic relationships among the *Striga*-resistant maize inbred lines than SSR markers, which is consistent with findings obtained in barley (Russell et al. 1997), coconut (Teulat et al. 2000), maize (Pejic et al. 1998) and olive (Belaj et al. 2003). The rapid and complex mutational processes within the SSR and flanking regions that result in size homoplasy (Peakall et al. 1998; Fisher et al. 2000) may underestimate the genetic diversity between more distantly related genotypes, which in turn may limit the use of SSRs for assessing relationships at the population levels (Peakall et al. 1998). Also, the high level of polymorphism detected with SSRs may reduce the statistical power to define relationships and thus lower the level of resolution attained with SSRs compared to other markers (Powell et al. 1996; Russell et al. 1997).

In summary, AFLPs and SSRs revealed a considerable level of genetic diversity among the *Striga*-resistant maize inbred lines, despite rigorous selection pressure imposed to recover lines with resistance to *Striga* from each source population. In our study, inbred lines originating from the same source population were genetically more similar to each other than those derived from different source populations, which is consistent with results from other studies (Ajmone-Marson et al. 1998; Mumm and Dudley 1994; Smith et al. 1997; Barbosa et al. 2003). The two marker types could not only distinguish between related inbred lines, but they also differentiated the lines on the basis of their genetic backgrounds. Considering the paucity of germplasm with good levels of resistance to *Striga* in maize, the broad genetic diversity detected with these two marker systems highlights the significant reservoir of diversity that exists among these lines for use in a breeding program. These results can aid in the selection and identification of potential sources of valuable genetic diversity to breed maize for resistance to *Striga*. The GS estimates can also be useful in breeding programs to maximize the level of variation in segregating populations by crossing lines with diverse genetic backgrounds. The high level of resistance fixed in these maize inbred lines can be exploited readily in breeding programs as parents of hybrids and synthetics and also as sources of favorable alleles to improve resistance to *Striga* in locally adapted germplasm.

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