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Inheritance of soybean resistance to soybean rust in Uganda's soybean germplasm

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Abstract

Understanding the genetic mechanisms of soybean rust resistance is important for effective selection and breeding procedures. This study was hence conducted to determine the combining ability and gene action controlling soybean rust using a 10x10 half diallel mating design. The F_2 segregating populations along with their parents were evaluated for rust severity and sporulation level at two reproductive stages (R4 and R6) in screen house and field conditions during the second season of 2016 and first season of 2017 at MUARIK using an alpha lattice design replicated thrice. Significant differences were observed among the parents and F_2 generations for both disease severity and sporulation level. General and specific combining abilities were highly significant. The GCA/SCA ratio (1.50-2.30) and the Baker's ratio (0.75-0.82) showed the predominance of additive gene action in the inheritance of soybean rust resistance. The broad-sense (0.94-0.99) and narrow-sense (0.73-0.82) heritability estimates indicated the possibility of improving resistance to soybean rust through selection in the early generations. UG 5, Maksoy 3N, Maksoy 4N and Maksoy 5N had negative GCA effects. The F_2 populations derived from these parents crossed with Wonder soya and Nam 2 had also negative SCA effects. The use of these parents and F_2 populations can, therefore, increase the response to selection for improving resistance to soybean rust.

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Introduction

Soybean, *Glycine max* (L.), is the world's most important legume in terms of production and trade due to its high protein (40%) and Oil (20%) content (Singh *et al.*, 2008). However, its yield in the tropical countries is low as compared to the temperate countries. The low yield in the tropics and, especially in Africa, is attributed to a number of abiotic and biotic factors (Singh and Rachie, 1987). Diseases, such as soybean cyst nematode, brown spot and soybean rust, are the most important among the biotic factors (Hartman *et al.*, 1999).

Soybean rust (SBR), caused by *Phakopsora pachyrhizi* (Sydow), is the most widespread and devastating foliar disease causing the greatest yield reduction in many tropical and sub-tropical areas of the world where soybean is cultivated and, hence, is considered a world threat to soybean production (Li *et al.*, 2012). The broad host range, high variability of virulence and rapid spread of *Phakopsora pachyrhizi* coupled with the potential of causing severe yield losses makes it to be the most destructive foliar disease of soybean (Hartman *et al.*, 2005). Moreover, the pathogen has a unique ability to directly penetrate the cuticle which accelerates its invasion capacity making its management a great challenge.

Phakopsora pachyrhizi is an obligate biotrophic plant-pathogenic fungus that colonizes leaf tissues (Miles *et al.*, 2006) forming small, water-soaked lesions that later develop into grey tan to dark brown or reddish brown (RB) lesions particularly on the abaxial leaf surfaces (Goellner *et al.*, 2010). The fungus causes substantial yield losses in different soybean growing areas of the world, particularly the sub-Saharan region of Africa (Kawuki *et al.*, 2003), the extent of which depends on susceptibility of the genotype, crop growth stage at which the disease starts, its intensity and weather conditions during the growing season (Hartman *et al.*, 2005).

Variable findings on the type of gene action and mode of inheritance of soybean resistance to rust have been reported by different genetic studies (Garcia *et al.*, 2008) as predominantly additive gene action (Maphosa *et al.*, 2012), partial and complete dominant gene action (Laperuta *et al.*, 2008), and epistatic gene action (Garcia *et al.*, 2008; Laperuta *et al.*, 2008). Ribeiro *et al.* (2009) also reported that additive effects predominated in the genetic control of soybean resistance to rust (*P. pachyrhizi*) and the interaction of the segregant populations with the environment did not alter the genetic parameter's general combining ability (GCA) and specific combining ability (SCA) estimates, indicating that estimates obtained in one year and one assessment can be extrapolated to others. These findings indicate that different selection procedures are needed to make genetic gains in rust resistance depending on the genotypes used, the environment under which the experiments are conducted and the durability of resistance being selected for.

In a diallel analysis for combining ability of self-pollinated crops like soybean, F_2 and F_3 generations were reported to give better predictions on the performance of lines due to the decreased level of dominance gene effects and availability of ample seed (Buhllar *et al.*, 1979). The objective of this study was, therefore: to determine the combining ability (GCA and SCA) effects in F_2 generations for soybean resistance to rust; and understand the heritability of the genes controlling resistance to soybean rust which would enhance effective selection and breeding procedures for future soybean improvement against the disease.

Materials and methods

Study site

This research was carried out at Makerere University Agricultural Research Institute-Kabanyolo (MUARIK) in screenhouse and field conditions during the second season of 2016 and first season of 2017. MUARIK is in Wakiso District of Central Uganda where severe seasonal soybean rust epidemics are frequent. It is located at an elevation of 1200 m above sea level (0°28'N and 32°37'E) and is 20 km north of Kampala, the capital city. The mean annual temperature at MUARIK ranges between 15-35.5°C with annual average rainfall of 332 mm and annual average humidity ranging between 13-96% (MUARIK Meteorology Station, 2017).

Progeny development and genotype evaluation

Ten soybean genotypes including three SBR susceptible cultivars (Kabanyolo I, Wondersoya and Nam 2), two moderately SBR resistant cultivars (Maksoy 1N and Namsoy 4M), three SBR resistant released varieties (Maksoy 3N, 4N and 5N), a high potential genotype (UG 5) with SBR resistance gene and a known source of SBR resistance PI 462312 (*Rpp3*) were used in the study whose details are shown in Table 1. Each genotype was planted in a screen-house in six plastic pots at different planting dates in order to synchronize flowering of varieties with different days-to-flowering. At flowering, all possible single crosses were made among the genotypes using a 10x10 diallel mating design with no reciprocals, following Griffing (1956) [Method 2/Model 1]. Out of the possible fifty-five cross combinations, fifty-three cross combinations were successfully generated. The resulting F_1 populations of each cross along with their corresponding parents were grown in plastic pots in the screenhouse and allowed to self-pollinate. True crosses were distinguished using morphological traits based on segregation and adequate F_2 and parental seeds were generated for subsequent field and screen-house evaluation.

Seeds from each parent and F_2 plants within a given cross were planted and evaluated for resistance to SBR under screen-house and field conditions at MUARIK. Both (field and screen-house) trials were laid in alpha lattice experimental design with three replications. All pots could not fit a single location (space) within the screenhouse. So, different areas of the screenhouse had to be used. Two meter rows containing 20-30 plants were used for each population, for the field experiment, with a spacing of 60 cm x 1.0 meter between rows and blocks, respectively. The distance within rows was 5 cm. To ensure high and uniform disease pressure in the plots, spreader rows of highly susceptible variety (Nam 2) was planted at the border rows surrounding the trial, according to the methodology described by Twizeyimana *et al.* (2007). In the screen-house, three pots (filled with soil from the field) per population with three plants per pot were used. The plants were inoculated with *P. Pachyrhizi* with a concentration of $5\text{x}10^4$ urediniospores per ml at V3 to V4 growth stages (about 3 weeks after sowing). Standard agronomic practices of weeding, fertilization, irrigation and pest control were applied during the entire season (Tukamuhabwa, 2006).

Data collection

Data on rust severity and sporulation level were collected from the parents and F_2 families at two reproductive growth stages (R4 – full pod stage and R6 – full seed stage). The data was scored from five randomly selected plants within a row (field) and three plants per pot (screen-house).

The disease severity and sporulation level scorings (Table 2) were taken using a 1- 5 scale as proposed by Miles *et al.* (2006). As SBR infections occur initially at the lower part of the canopy and progress upwards (Kumudini *et al.*, 2008), the scoring from each of the sample plants was divided into three canopy sections (bottom, middle and top) (Kawuki *et al.*, 2004). Rust severity and sporulation level of the entire plant were determined based on the average score of the three canopy levels. Similarly, the resistance level of the genotype was determined based on the average score of the sampled five plants and three plants for the field and screenhouse experiments, respectively. Reaction responses were assessed and grouped into reddish brown (RB), tan (T) and mixed reactions (M) at R6 growth stage, which coincides with the period when soybean plants are severely infected by SBR (Kawuki *et al.*, 2004).

Data analysis

The collected data for the traits measured were analysed using linear mixed model (LMM), appropriate method for alpha-lattice design, in GENSTAT statistical package 16th edition (Payne *et al.*, 2013).

The mixed model used was:

$$\boxed{Y_{ijk} = \mu + G_i + R_j + R/B_{j(k)} + \varepsilon_{ijk}}$$

Where, Y_{ijk} = observed value from each experimental unit, μ = population mean, G_i = effect of i^{th} genotype, R_j = effect of the j^{th} replicate, $R/B_{k(j)}$ = effect of the k^{th} block nested to the j^{th} replicate, and ε_{ijk} = the experimental error. Genotype effect was considered as fixed while block and replication effects as random.

The general combining ability (GCA) of the parental genotypes and the specific combining ability (SCA) for the F_2 generation crosses were estimated using the Griffing's Method 2/Model 1 (Griffing, 1956). Student's t-test was applied to examine the significance of the general and specific combining ability of the traits.

The gene action and the relative importance of GCA and SCA were determined using the general predicted ratio (GPR) for all the traits as illustrated by Baker (1978).

The additive and dominance variances and heritability were estimated according to Singh and Chaudhary (2004). The Baker's ratio (BR), narrow sense (h^2) and broad sense (H^2) heritability were calculated as follows:

$$BR = \frac{2VCg_i}{2VCg_i + VCs_{ij}};\quad h^2 = \frac{2VCg_i}{2VCg_i + VCs_{ij} + MS_{Lee}};\quad H^2 = \frac{2VCg_i + VCs_{ij}}{2VCg_i + VCs_{ij} + MS_{Lee}}$$

Where, VCg_i= the variance component for the GCA effect of the ith parent, VCs_{ij}= the variance component for the SCA effect of the ijth cross (i x j), MS_{Lee}= the mean square of lattice effective error.

Results

The analysis of variance (ANOVA) for soybean rust severity and sporulation level at R4 and R6 growth stages showed highly significant difference ($P \leq 0.001$) among the parents and segregating F_2 generations both under field and screen-house conditions (Table 3).

Means of disease severity and sporulation level at both growth stages as well as lesion types are shown in Table 4. In both field and screenhouse, the highest scores were recorded at R6. For the screenhouse, UG 5 had the least mean rust severity (R4=1.63, R6 = 2.11) and sporulation level (R4 = 1.30; R6 = 1.63). In the field, the lowest mean rust severity was recorded from the parental genotype Maksoy 3N (R4 = 1.18) and the cross Maksoy 3N x Maksoy 4N (R6 = 1.53), while the lowest mean sporulation level was recorded from the cross Maksoy 3N x UG 5 (R4 = 0.99) and the parental genotype Maksoy 3N (R6 = 1.37).

Table 1. Description of ten parental genotypes used in the half-diallel mating design at MUARIK during the second season of 2016 and first season of 2017.

Parental lines	Pedigree	Origin/Source	Response to rust
Maksoy 3N	Duiker x TG x 1835-10E	MAK, Uganda	Resistant
Maksoy 4N	Duiker x GC00138-29	MAK, Uganda	Resistant
Maksoy 5N	Nam II × GC00138-29	MAK, Uganda	Resistant
UG 5	-	MAK, Uganda	Resistant
PI 462312 (Rpp3)	G7955	AVRDC, Taiwan	Resistant (Rpp3)
Maksoy 1N	TGx1835-10E	MAK, Uganda	Moderately resistant
Namsoy 4M	Nam 2 × GC00138-29	NARO, Uganda	Moderately resistant
Kabanyolo I	Mutant of Clark 63	Uganda	Susceptible
Nam 2	87D-668	Nigeria	Susceptible
Wondersoya	-	IITA, Nigeria	Susceptible

MAK: Makerere University; AVRRDC: The World Vegetable Development Centre; NARO: National Agricultural Research Organization, Uganda; IITA: International Institute for Tropical Agriculture (Source: Maphosa et al., 2012).

The parental genotypes Wondersoya and Nam 2, and their cross (Wondersoya x Nam 2) were the most susceptible with rust severity ranging between 3.87-4.48 and sporulation level ranging between 3.6-4.15.

With regard to the lesion type, TAN, reddish brown (RB) and mixed (M) lesions were observed.

UG 5 and Maksoy (3N, 4N and 5N) parental genotypes and their F_2 progenies showed reddish brown (RB) lesions while PI 462312 (*Rpp3*) showed TAN lesions under both field and screenhouse conditions. Mixed (M) lesions were observed in some parents and F_2 generations.

Table 2. Description of 1 - 5 scale for soybean rust assessment.

Score	Description	
	Disease severity	Sporulation level
1	No visible lesions	No sporulation
2	Few scattered lesions present	<25% of fully sporulating lesions
3	Moderate number of lesions on at least part of the leaf	26-50% of fully sporulating lesions
4	Abundant number of lesions on at least part of the leaf	51-75% of fully sporulating lesions
5	Prolific lesions development over most of the leaf	Fully sporulating TAN lesions

Source: Miles *et al.* (2006).

The mean squares ANOVA of general combining ability (GCA) and specific combing ability (SCA) are shown in Table 5 and revealed highly significant variations for disease severity and sporulation level among the ten parental genotypes and 43 segregating F_2 generations, respectively. The GCA/SCA ratio was greater than one (1.50 – 2.30), the Baker's ratio (BR) was high (0.75 – 0.82) and the heritability estimates were high, 0.94 – 0.99 (broad-sense) and 0.73 – 0.81 (narrow sense), for both disease severity and sporulation level at both growth stages and sites.

Estimates of GCA effects for rust severity and sporulation level showed highly significant variation ($P \leq 0.001$) among individual parents (Table 6). At R6, GCA effects for rust severity ranged from -0.527–0.591 (field) and -0.549–0.527 (screenhouse), while for sporulation level ranged from -0.512–0.589 (field) and -0.624–0.565 (screenhouse).

Table 3. Means and mean squares of disease severity and sporulation level.

Source	d.f.	Disease Severity (DS)				Sporulation Level (SL)			
		Field		Screenhouse		Field		Screenhouse	
		R4	R6	R4	R6	R4	R6	R4	R6
Gen	52	0.425^{***}	1.325^{***}	0.942^{***}	1.321^{***}	0.423^{***}	1.378^{***}	1.215^{***}	1.744^{***}
Rep	2	0.286^{***}	0.286^{***}	0.928^{***}	0.671^{***}	0.301^{***}	0.213^{**}	1.100^{***}	0.799^{***}
Rep. Block	20,21	0.049^{*}	0.033^{ns}	0.009^{*}	0.020^{*}	0.052^{*}	0.026^{ns}	0.008^{*}	0.013^{ns}
Lee	74,75	0.027	0.035	0.014	0.018	0.029	0.033	0.018	0.016
Mean		1.60	2.32	2.34	2.87	1.43	2.05	2.03	2.51
s.e.		0.0424	0.0424	0.0765	0.0834	0.0436	0.0366	0.0834	0.0709
CV (%)		10.22	8.06	4.99	4.73	12.03	8.85	6.52	5.06

R4=full-pod stage; R6=full-seed stage; d.f.=degree of freedom; s.e.=standard error; *,** and ***=significant at $P \leq 0.05, 0.01$ and 0.001, respectively; ns=non significant at $P \leq 0.05$.

Among the parents, Maksoy (3N, 4N, 5N) and UG 5 were the best general combiners with a highly significant ($P \leq 0.001$) and negative GCA effects. On the other hand, Wondersoya and Nam 2 consistently exhibited the highest GCA effects with highly significant ($P \leq 0.001$) and positive values for rust severity and sporulation level followed by Kabanyolo I both at field and screenhouse conditions.

Inconsistent results of GCA effects were observed on Namsoy 4M and PI 462313 (*Rpp3*). Under screenhouse condition, these parents showed highly significant and positive GCA effects for rust severity and sporulation level at both reproductive stages. However, under field condition, PI 462313 (*Rpp3*) showed non-significant GCA effects for rust severity and sporulation level at R4 while Namsoy 4M showed non-significant GCA effects for rust severity (R4 and R6) and sporulation level (R4).

Table 4. Mean scores of soybean rust severity, sporulation level and lesion type of some representative parental and F_2 populations at field and screenhouse (MUARIK).

Population	Disease severity				Sporulation level				Lesion type
	Field		Screenhouse		Field		Screenhouse		
	R4	R6	R4	R6	R4	R6	R4	R6	
Maksoy 3N x Maksoy 4N	1.20	1.53	1.81	2.26	1.09	1.38	1.48	1.78	RB
Maksoy 3N	1.18	1.62	1.70	2.19	1.05	1.37	1.37	1.74	RB
UG 5	1.20	1.62	1.63	2.11	1.06	1.42	1.30	1.63	RB
Maksoy 1N x Maksoy 3N	1.24	1.69	1.86	2.41	1.09	1.44	1.49	1.89	RB
Maksoy 3N x UG 5	1.19	1.69	1.70	2.19	0.99	1.38	1.33	1.70	RB
Maksoy 1N x UG 5	1.36	1.70	1.89	2.30	1.27	1.47	1.55	1.81	RB
Maksoy 3N x Rpp3	1.29	1.76	1.89	2.33	1.17	1.51	1.56	1.96	RB
Namsoy 4M x UG 5	1.24	1.82	2.04	2.37	1.07	1.51	1.70	2.00	RB
Wondersoya x Maksoy 5N	1.51	2.02	2.15	2.59	1.36	1.73	1.81	2.22	M
Namsoy 4M x Rpp3	1.41	2.14	2.89	3.44	1.24	1.93	2.67	3.22	M
Namsoy 4M x Maksoy 1N	1.86	2.51	2.67	3.26	1.73	2.29	2.37	3.00	M
Namsoy 4M	1.73	2.58	2.85	3.19	1.56	2.42	2.63	3.00	M
Wondersoya x Maksoy 1N	2.11	2.98	3.00	3.56	2.00	2.67	2.78	3.37	M
Maksoy 1N	1.75	3.07	2.89	3.52	1.58	2.67	2.59	3.26	M
Nam 2 x Namsoy 4M	2.12	3.16	2.89	3.67	1.93	2.89	2.67	3.44	M
PI 462312 (Rpp3)	2.10	3.07	2.70	3.48	1.89	2.76	2.44	3.19	T
Wondersoya x Kabanyolo I	2.37	3.56	3.11	3.74	2.18	3.18	2.9	3.6	T
Nam 2	2.52	3.87	3.26	4.22	2.33	3.60	3.04	3.85	T
Wondersoya x Nam 2	2.69	4.02	3.45	4.37	2.56	3.80	3.27	4.11	T
Wondersoya	2.76	4.20	3.48	4.48	2.56	3.98	3.11	4.15	T
L.S.D.	0.29	0.3	0.18	0.22	0.30	0.29	0.20	0.20	

R4=full pod-stage; R6=full-seed stage; T=tan coloured lesions; RB=reddish brown lesions; M=mixed lesions; l.s.d.=least significance difference.

Estimates of SCA effects for rust severity and sporulation level for some representative crosses are presented in Table 7 for both field and screenhouse conditions.

The F_2 progeny of crosses Wondersoya x Maksoy 3N and Nam 2 x Maksoy 3N consistently showed highly significant negative SCA effects ($P \leq 0.001$) for both severity and sporulation level at both field and screenhouse conditions. At R6 growth stage, the F_2 progeny of cross Wondersoya x Maksoy 4N showed the least SCA effects for rust severity (-0.531 under field condition) and sporulation level (-0.538 and -0.548, under field and screenhouse conditions, respectively), while the cross Wondersoya x UG 5 showed the least SCA effect (-0.478) for rust severity under screenhouse condition.

Inconsistent SCA effects were observed for the F_2 progeny crosses involving PI 462312. For example, Wondersoya x PI 462312 showed significant and negative SCA effects for disease severity (R4 and R6) and sporulation level (R4) under field condition while significant and positive SCA effects for sporulation level (R4 and R6) and disease severity (R6) under screenhouse condition. Similarly, Nam 2 x PI 462312 showed significant and positive SCA effects for rust severity and sporulation level under screenhouse but non-significant SCA effects under field condition.

Discussion

The existence of variation for rust severity and sporulation among the parents and segregating F_2 generations is an essential condition for studying the genetic control of soybean rust resistance trait.

Soybean rust severity and sporulation level were significantly variable at both growth stages with the highest scores recorded at R6 indicating that R6 is the stage where soybean plants are most severely infected by SBR (Kawuki *et al.*, 2004).

Table 5. Mean squares of disease severity and sporulation level for GCA and SCA in ten parental genotypes and 43 F_2s under field and screenhouse condition.

Source	d.f.	Disease severity (DS)				Sporulation level (SL)			
		Field		Screenhouse		Field		Screenhouse	
		R4	R6	R4	R6	R4	R6	R4	R6
Crosses	52	0.167***	0.440***	0.293***	0.452***	0.165***	0.434***	0.340***	0.566***
GCA	9	0.747***	1.958***	1.411***	2.095***	0.733***	1.923***	1.639***	2.662***
SCA	43	0.046***	0.122***	0.058***	0.108***	0.046***	0.122***	0.068***	0.128***
Lee	104	0.009***	0.012***	0.005***	0.006***	0.010***	0.011***	0.006***	0.005***
VCgca/VCsca		1.74	1.54	2.28	1.80	1.73	1.50	2.30	1.89
BR		0.78	0.75	0.82	0.78	0.78	0.75	0.82	0.79
h^2		0.74	0.74	0.81	0.77	0.73	0.73	0.81	0.78
H^2		0.95	0.97	0.99	0.99	0.94	0.98	0.98	0.99

R4=full pod-stage; R6=full seed-stage; d.f.=degree of freedom; GPR.=Baker's ratio; h^2=narrow sense heritability; H^2=broad sense heritability; Lee=lattice effective error; ***=significant at $P \leq 0.001$.

Moreover, R6 is the growth stage where clear differences are observed between susceptible and resistant genotypes thereby indicating the overall susceptibility or resistance level of a genotype (Kawuki *et al.*, 2004). The highest severity and sporulation mean scores were also recorded from the screenhouse as compared to the field condition in all the parental and F_2 segregating populations. This could be due to favorable conditions (high humidity, moderate temperature and prolonged leaf wetness) created in the screenhouse for the development of the pathogen (Bromfield, 1984).

Table 6. Estimates of GCA effects of ten parental genotypes for rust severity and sporulation level tested in field and screenhouse.

F_2 Genotypes	Field Condition				Screenhouse Condition			
	DS_R4	DS_R6	SL_R4	SL_R6	DS_R4	DS_R6	SL_R4	SL_R6
Wondersoya	0.380	0.591	0.378	0.589	0.405	0.527	0.425	0.565
Nam 2	0.368	0.575	0.357	0.568	0.353	0.480	0.386	0.504
Kabanyolo-I	0.194	0.270	0.191	0.251	0.235	0.257	0.258	0.298
Namsoy 4M	0.014^{ns}	0.045^{ns}	0.022^{ns}	0.061*	0.195	0.148	0.220	0.216
Maksoy 1N	0.067**	0.171	0.081**	0.161	0.197	0.232	0.202	0.261
Maksoy 3N	-0.325	-0.527	-0.313	-0.512	-0.465	-0.519	-0.500	-0.622
Maksoy 4N	-0.269	-0.451	-0.265	-0.431	-0.412	-0.481	-0.441	-0.531
Maksoy 5N	-0.254	-0.366	-0.256	-0.382	-0.288	-0.392	-0.319	-0.434
UG 5	-0.219	-0.427	-0.231	-0.439	-0.448	-0.549	-0.482	-0.624
PI 462312	0.041^{ns}	0.113	0.034^{ns}	0.127	0.212	0.281	0.236	0.345

DS_R4=disease severity at full-pod stage; DS_R6=disease severity at full-seed stage; SL_R4=sporulation level at full-pod stage; SL_R6=sporulation level at full-seed stage; **,*=significant at $P \leq 0.01$ and 0.05, respectively; ns=non-significant at $P=0.05$; the rest are significantly different at $P \leq 0.001$.

The local genotype UG 5 and the three improved varieties (Maksoy 3N, 4N and 5N) had lower mean score values and were resistant to soybean rust; while Wondersoya and Nam 2 had higher mean score values indicating their susceptibility to soybean rust. However, the parental genotype PI 462312, with the known resistance gene *Rpp3*, showed moderate (at R4) to high (at R6) mean score values for severity and sporulation indicating possible breakdown of resistance. When evaluated at R6 reproductive stage, the parental genotypes and F_2 populations with low mean severity and sporulation level showed RB lesions while those with high mean severity and sporulation level showed TAN lesions.

Similarly, the parent PI 462312 (*Rpp3*) showed TAN lesions under both field and screenhouse conditions. This result agrees to the findings of Miles *et al.* (2011) and Souza *et al.* (2014) that *Rpp3* resistance was ineffective and showed TAN lesions. However, contradicting findings by Godfrey (2015), Maphosa *et al.* (2013) and Li (2009) showed RB lesions and resistance to soybean rust by the parent PI 462312.

Table 7. Estimates of SCA effects of rust severity and sporulation level for some representative crosses under field and screenhouse conditions.

F_2 Genotype	Field condition				Screenhouse condition			
	DS_R4	DS_R6	SL_R4	SL_R6	DS_R4	DS_R6	SL_R4	SL_R6
W x N-II	0.348^{***}	0.532^{***}	0.396^{***}	0.596^{***}	0.353^{***}	0.491^{***}	0.427^{***}	0.531^{***}
W x K-I	0.198^{*}	0.372^{***}	0.185^{*}	0.291^{**}	0.139^{*}	0.084^{ns}	0.185^{**}	0.218^{**}
W x 4M	-0.033^{ns}	0.018^{ns}	-0.024^{ns}	-0.075^{ns}	0.027^{ns}	-0.026^{ns}	0.065^{ns}	0.000^{ns}
W x 1N	0.068^{ns}	-0.108^{ns}	0.117^{ns}	-0.130^{ns}	0.063^{ns}	-0.074^{ns}	0.123^{ns}	0.030^{ns}
W x 3N	-0.298^{***}	-0.431^{***}	-0.312^{***}	-0.480^{***}	-0.347^{***}	-0.399^{***}	-0.353^{***}	-0.529^{***}
W x 4N	-0.295^{***}	-0.531^{***}	-0.293^{**}	-0.538^{***}	-0.402^{***}	-0.472^{***}	-0.421^{***}	-0.548^{***}
W x 5N	-0.214^{*}	-0.526^{***}	-0.190^{*}	-0.521^{***}	-0.304^{***}	-0.413^{***}	-0.320^{***}	-0.423^{***}
W x UG 5	-0.193^{*}	-0.466^{***}	-0.239^{**}	-0.508^{***}	-0.323^{***}	-0.478^{***}	-0.333^{***}	-0.491^{***}
W x Rpp3	-0.383^{***}	-0.250^{*}	-0.391^{***}	-0.142^{ns}	0.122^{ns}	0.173^{*}	0.163^{*}	0.205^{***}
N-II x 4M	0.146^{ns}	0.212^{*}	0.129^{ns}	0.212^{*}	0.006^{ns}	0.169^{*}	0.031^{ns}	0.211^{**}
N-II x 1N	0.089^{ns}	0.197^{ns}	0.070^{ns}	0.224^{*}	0.108^{ns}	0.086^{ns}	0.074^{ns}	0.125^{ns}
N-II x 3N	-0.361^{***}	-0.462^{***}	-0.396^{***}	-0.483^{***}	-0.260^{***}	-0.460^{***}	-0.285^{***}	-0.507^{***}
N-II x 4N	-0.238^{**}	-0.448^{***}	-0.228^{*}	-0.451^{***}	-0.276^{***}	-0.461^{***}	-0.308^{***}	-0.413^{***}
N-II x 5N	-0.199^{*}	-0.489^{***}	-0.170^{ns}	-0.477^{***}	-0.289^{***}	-0.402^{***}	-0.319^{***}	-0.472^{***}
N-II x UG 5	-0.166^{ns}	-0.361^{***}	-0.216^{*}	-0.400^{***}	-0.284^{***}	-0.392^{***}	-0.316^{***}	-0.397^{***}
N-II x Rpp3	-0.001^{ns}	0.032^{ns}	0.027^{ns}	-0.053^{ns}	0.204^{***}	0.185^{*}	0.225^{**}	0.263^{***}

DS_R4=disease severity at full-pod stage; DS_R6=disease severity at full-seed stage; SL_R4=sporulation level at full-pod stage; SL_R6=sporulation level at full-seed stage; *, ** & ***=significant at P≤0.05, 0.01 & 0.001; ns=non-significant at P=0.05.

The breakdown of resistance to soybean rust in this genotype could, therefore, be due to the high degree of virulence nature of the pathogen as reported by Tukamuhabwa *et al.* (2009) and Shan *et al.* (2012). In previous studies, host-pathogen interaction that resulted in RB reactions tended to have longer latent periods, lower rates of increase in pustule number over time and smaller lesions which are characteristics of partial resistance (Walker *et al.*, 2011) while the susceptible interactions resulted in TAN reaction (Bromfield, 1984; Miles *et al.*, 2006).

Mixed reactions (M), RB and TAN lesions on the same plant, were also observed in some of the parents and F_2 populations indicating the heterogeneity of the pathogen population at MUARIK. Similar findings were reported by previous studies (Miles *et al.*, 2008; Maphosa *et al.*, 2012, 2013).

Highly significant variations for GCA and SCA mean squares suggested the importance of additive and non-additive gene effects in determining the inheritance of resistance to SBR (Dabholkar, 1937).

High Baker's ratio (BR) and greater than one GCA/SCA ratio revealed the relative importance of GCA effect with respect to SCA effect (Baker, 1978) indicating the predominance of additive gene action in the inheritance of soybean resistance to rust. A study from an F_2 population generated from a half diallel mating design also revealed that additive gene action played a significant role in the inheritance of rust resistance (Wanderi, 2012). Similar results were reported by Ribeiro *et al.*, (2009), Maphosa *et al.* (2012) and Martins and Juliatti (2014). The high broad-sense heritability (H^2) estimate indicated that high proportion of the phenotypic variation is due to genetic effect. The high narrow-sense heritability (h^2) suggested that selection for resistance to soybean rust can be done at early stages, because high additive gene action and heritability lead to high response to selection (Acquaah, 2012).

The significant difference observed for GCA effects among the ten parental genotypes indicated the variation in the level of contribution of the genotypes to rust resistance. UG 5 and Maksoy (3N, 4N and 5N) showed highly significant negative GCA values indicating a larger contribution towards resistance and hence are desirable for rust resistance (Lokko *et al.*, 2004).

These parents are, therefore, best general combiners for soybean resistance to rust and their use in breeding programs would produce progenies with increased resistance. However, Wondersoya, Nam 2, Kabanyolo I and Maksoy 1N showed highly significant positive GCA values. These parents contributed least to SBR resistance and hence their progenies would show increased susceptibility to soybean rust if used in hybridization programs (Lokko *et al.*, 2004). The parents with inconsistent GCA effects, PI 462312 (*Rpp3*) and Namsoy 4M, are not useful to generate variability for selecting lines with resistance to soybean rust.

Specific combining ability (SCA) is used to determine combinations which do relatively better or worse than would be expected on the basis of average performance of lines involved (Sprague and Tatum, 1942). In this study, negative SCA effects were desirable for resistance. Estimates of SCA effects were significantly variable among the F_2 segregating populations. In general, significant and negative SCA effects were observed in all the crosses involving parents with contradicting extremes of resistance (highly susceptible and highly resistance), with the exception of Nam 2 x UG5 and Nam 2 x Maksoy 5N which showed negative but non-significant SCA effects for rust severity and sporulation, respectively, at R4 stage and field condition. In self-pollinated crops like soybean, desirable SCA or non-additive effect suggests that it is possible to select superior plants in the progenies of a cross for a trait of interest.

Conclusion

The local genotype UG 5 and the three improved varieties were found to be resistant to SBR while the parental genotype PI 462312 with the known resistance gene *Rpp3* was found to be susceptible to SBR. Low rust severity and sporulation level were observed from F_2 populations of parents with high negative GCA effects for resistance to rust such as Maksoy (3N, 4N, 5N) and UG 5 at both reproductive stages and different environmental conditions. The significant differences observed for GCA and SCA for rust severity and sporulation levels suggested the role of additive and non-additive genetic effects in controlling resistance to rust. Greater than unity GCA/SCA ratios and high baker's ratios (BR) indicated the predominance of additive gene action over dominance for soybean resistance to rust.

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