EVALUATION OF COWPEA GENOTYPES FOR THEIR REACTIONS TO STEM ROT INDUCED BY *RHIZOCTONIA SOLANI* (KUHN)

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SUMMARY

Rhizoctonia solani is one of the pathogens that infect cowpea causing stem and root rot, seedling decay and damping - off complex which result in low yield. This study aims to evaluate some cowpea genotypes for their reactions to *Rhizoctonia solani.*

Six improved cowpea varieties (SAMPEA 7, SAMPEA 8, SAMPEA 9, SAMPEA 10, SAMPEA 11 and SAMPEA 12) two accessions (IAR–00–1074 and IAR–07–1050) and four local varieties (Biu local, Maifarinhanci, Kanannado and Dansokoto) were evaluated in the glasshouse and on the field for their reaction to *R. solani* infection. *R. solani* was multiplied on millet grain and used to inoculate the cowpea genotypes in both glasshouse and field studies. Symptoms of disease incidence and severity were observed on all the 12 cowpea genotypes inoculated with *R. solani.* In the glasshouse, SAMPEA 10 recorded the highest incidence (100 %) and SAMPEA 9 recorded the highest severity (62.86 %). In the field however SAMPEA 10 recorded the highest incidence (75.41 %) and severity (66.19 %). Dansokoto recorded the lowest incidence (8.33 and 21.67 % for glasshouse and field respectively) and severity (6.67 and 17.14 % for glasshouse and field respectively). Correlation analysis showed that stem rot incidence and severity significantly resulted in reduced number of pods and grain yield. It is thus concluded that the local varieties evaluated are more resistant to the disease and as such breeding program should focus on improving the desired characteristics of the local varieties.

Keywords: Screening, stem rot, cowpea, genotypes, fungi

Stem rot constitute a constraint to cowpea production in Nigeria (16). was reported in Sri Lanka (5), Tanzania, Benin, South Africa (2) and Nigeria (14, 16) to be caused by a complex of fungi including species of *Pythium*, *Phytophthora*, *Sclerotium*, *Rhizoctonia*, *Macrophomina* and *Fusarium*. Stem rot is characterized by reddish brown lesions on the stem at soil level which may extend upward to about the fifth node but rarely to the growing tip. The lesions may girdle the stem at the first node or a little below it. The leaf petioles at the nodes and the basal portion of some side shoots are also invaded by the fungus. Yield loss of 11 – 40 % caused by stem rot induced by *Pythium aphanidermatum*, S*clerotium rolfsii* and *Fusarium solani* at Moor plantation, Ibadan was reported (14). According to Isubikalu *et al*. (6), most farmers have some knowledge on methods of controlling insect pests but have little knowledge of controlling cowpea diseases (6). The most frequently used control measure against pests and diseases is by synthetic pesticides usage whose intensive and indiscriminate use in agriculture has caused many problems to the environment. Such problems include water, soil, animals and food contamination; poisoning of farmers; The disease

elimination of non-target organisms; and selection of pest and weed tolerant to certain pesticides (11). Alternative control measures of plant diseases include cultural control, biological control, regulatory control and the use of crops resistant to certain pests and diseases. The use of resistant crop varieties is the most economical, safest and one of the most effective means of managing plant diseases in crops (12). It is therefore important for scientists to give appropriate research attention to screening and breeding cowpea varieties resistant to stem rot to minimize losses caused by the disease in a manner that will not pose threat to the environment, human and beneficial animals. This study aims to evaluate some cowpea varieties reaction to stem rot so as to serve as a source of materials towards breeding for resistance.

MATERIALS AND METHODS

The experiment was conducted in Samaru Zaria (11° 12′ N 07° 37′ E.). Six improved cowpea varieties (SAMPEA 7, SAMPEA 8, SAMPEA 9, SAMPEA 10, SAMPEA 11 and SAMPEA 12), two accessions (IAR–00–1074 and IAR–07–1050) and four local varieties (Biu local, Maifarinhanci, Kanannado and Dansokoto) were obtained from the Legumes and oil seed programme of Institute for Agricultural Research (IAR), Ahmadu Bello University Zaria.

The culture of *Rhizoctonia solani* used was obtained by isolation from diseased cowpea stem using standard procedure of disinfecting and plating on Potato Dextrose Agar (PDA). The fungus was identified using cultural and microscopic characteristics based on the manual by Watanabe (18). Identity of the isolate was confirmed by the Centre for Biosciences and Agriculture International (CABI), United Kingdom.

Glasshouse Evaluation

The cowpea genotypes were evaluated for their reactions to the causal organism in the glasshouse. Seeds were sterilized in 1 % Sodium hypochlorite (NaOCl) for 2 minutes, rinsed twice in SDW and sown at three seeds per pot. Pots were arranged in Completely Randomised Design with four repetitions. Each genotype constitutes a treatment and is planted in two pots per repetition. The plants were inoculated with *Rhizoctonia solani* growing on millet grain (50 g/pot) one week after sowing (WAS). Disease incidence and severity on the plants were recorded weekly at 2, 4 and 6 WAS. Disease incidence was calculated as percentage of diseased plants in each repetition.

Individual plants were scored for disease severity based on a modified scale of 0 - 7 by Fernando and Linderman (5) where

0 = No visible symptoms.

1 = Lesions at base of stem (≤ 15 % stem length); leaves pale green

2 = Lesions advanced upward (16 - 30 % stem length); leaves wilting

3 = Lesions advanced upward (31 - 45 % stem length); leaves wilted

4 = Lesions advanced upward (46 - 60 % stem length); stem rotting at base

5 = Lesions advanced upward (> 60 % stem length); stem rotten at base

6 = Plant rotten, wilted and death

Disease severity percentage was determined using the following formula

$$\text{Disease severity (\%)} = \frac{\text{Sum of disease rating}}{\text{Total number of plants rated} \times \text{Highest rating}} \times 100$$

Using the severity values, the genotypes were grouped into five reaction groups based on a modified scale of Adejumo *et al*. (3) where varieties with severities (%) of

0 = Immune

1 - 15 = Highly resistant

16 – 30 = Moderately resistant

31 – 45 = Moderately susceptible

> 45 = Highly susceptible

Pod number and grain yield were recorded for each repetition and expressed per plant.

Field Evaluation

The 12 cowpea genotypes were also evaluated under field conditions in 2014. The genotypes were arranged in a Randomized Complete Block Design (RCBD) with three replications. In a replicate, each variety was sowed on 12m^2 plot using three seeds per hole and an intra and inter row spacing of 20 cm and 7 cm were maintained. Plots were separated from each other by one bare, 75 cm weeded ridge while replicates were separated from each other by 1 m wide margin. One week after sowing, the top soil of each stand was inoculated by incorporating 50 g of millet seed inoculum while sterilized millet seeds were incorporated in control plots. Disease incidence and severity on 6.0 m^2 net plots were recorded at 2, 4 and 6 WAS. Cowpea pods were harvested at maturity and data on pod number per plot was collected at harvest and grain yield per plot was obtained after threshing and expressed per hectare. Varieties were separated into different resistant groups using the scale described above

Statistical Analysis

The data generated were subjected to analysis of variance (ANOVA) procedure of SAS and means were separated using the Duncan Multiple Range Test (DMRT) at 5 % level of significance.

RESULTS

Disease incidence and severity in the glasshouse

The incidence of stem rot on cowpea genotypes inoculated with *Rhizoctonia solani* in the glasshouse significantly varied (Table 1). At 2 Weeks after Sowing (WAS), highest disease incidence was recorded on SAMPEA 10 which did not differ significantly from that of Biu local and IAR-07-1050. Dansokoto, IAR-00-1074, SAMPEA 11 and SAMPEA 12 were disease free.

At 4 WAS, Biu local and SAMPEA 10 recorded the highest incidence (91.67 %) which did not differ significantly from that of IAR- 07-1050. Dansokoto recorded the lowest incidence (7.5 %) which did not significantly differ from that of IAR-00- 1074, SAMPEA 8 and SAMPEA 11

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Table 1: Disease incidence and severity of stem rot on cowpea genotypes inoculated with *Rhizoctonia solani* in the glasshouse

Variety	Disease incidence (%)			Disease severity		
	2WAS	4WAS	6WAS	2WAS	4WAS	6WAS
SAMPEA 10	75.00^{a}	91.67^{a}	100.00^{a}	29.5	32.38^{b}	55.71^{b}
IAR- 07- 1050	70.83^{a}	87.50^{a}	100.00^{a}	0.00^{f}	11.43^{fg}	17.14^{ef}
Biu local	54.17^{a}	91.67^{a}	100.00^{a}	0.00^{f}	20.00^{c-f}	28.53^{cde}
SAMPEA 8	23.33^{b}	15.00^{cd}	33.33^{cd}	35.24^{ab}	55.24^{a}	60.95^{a}
SAMPEA 9	20.83^{b}	45.83^{b}	79.17^{b}	40.76^{a}	58.09^{a}	62.86^{a}
Kanannado	15.00^{b}	37.50^{bc}	37.50^{cd}	6.67^{ef}	20.95^{b-e}	37.14b^{cd}
SAMPEA 7	8.33^{b}	30.17^{bcd}	45.83^{cd}	19.05^{cd}	13.33^{d-g}	24.76^{cde}
Maifarinhanci	7.50^{b}	33.33^{bc}	33.33^{cd}	19.05^{cd}	24.76^{bcd}	39.81^{bc}
SAMPEA 11	0.00^{b}	15.83^{cd}	27.50^{de}	0.00^{f}	12.38^{efg}	22.86^{de}
SAMPEA 12	0.00^{b}	37.50^{bc}	50.00c	5.71^{ef}	31.43^{bc}	31.43^{b-e}
IAR- 00- 1074	0.00^{b}	15.00^{cd}	27.50^{de}	11.43^{de}	23.81^{b-e}	23.81^{de}
Dansokoto	0.00^{b}	7.50^{d}	8.33^{e}	0.00^{f}	5.71^{g}	6.67^{f}
SE±	7.47	7.62	6.29	3.53	3.82	4.88

Means followed by similar superscript in a column are not significantly different at 5 % level of significance ($P \leq 0.05$) using Duncan Multiple Range Test (DMRT)

WAS = Weeks after sowing

At 6 WAS, IAR-07-1050, Biu local and SAMPEA 10 recorded the highest incidence (100 %) followed by SAMPEA 9. Dansokoto recorded the lowest incidence (8.33 %) which did not significantly differ from that of IAR-00-1074 and SAMPEA 11.

The severity of stem rot on cowpea genotypes inoculated with *R. solani* in the glasshouse was significant (Table 1). At 2 WAS; highest severity (40.76 %) was recorded on SAMPEA 9 and lowest severity (0.00 %) were from IAR-07-1050; Biu local; SAMPEA 7; SAMPEA 11 and Dansokoto. This trend was similar at 4 WAS and at 6 WAS, Dansokoto had the lowest severity (6.67 %) and this was significantly lower than SAMPEA 11; SAMPEA 7 and Biu Local but not IAR-07-1050 at 17.4 %.

Disease incidence and severity on the field

The incidence of stem rot on cowpea genotypes inoculated with *R. solani* on the field also significantly varied (Table 2). At 2 WAS, highest disease incidence was recorded on SAMPEA 10 (14.79 %) which did not differ significantly from that of IAR-00-1074, SAMPEA 12, IAR-07-1050 and Biu local. SAMPEA 7, SAMPEA 8, SAMPEA 11, Dansokoto, Kanannado, Maifarinhanci and SAMPEA 9 were disease free.

At 4 WAS, SAMPEA 11 recorded the highest incidence (42.94 %) which did not differ significantly from that of SAMPEA 10, Maifarinhanci and SAMPEA 9. Dansokoto recorded the lowest incidence (21.67 %) which did not differ significantly from that of SAMPEA 8, IAR-00-1074, Kanannado, IAR-07-1050, Biu local, SAMPEA 7 and SAMPEA 9. At 6 WAS, SAMPEA 10 recorded the highest incidence (75.41 %) followed by Biu local which did not significantly differ from that of IAR-07-1050. Dansokoto recorded the lowest incidence (21.67 %) and was not significantly different from that of Kanannado (25.29 %) and SAMPEA 12 (30.28 %).

The severity of stem rot on cowpea genotypes inoculated with *R. solani* on the field also significantly varied (Table 2). At 2 WAS, highest disease severity (9.99 %) was recorded on SAMPEA 10 while Dansokoto, Kanannado, and Maifarinhanci were disease free.

At 4 WAS, SAMPEA 11 recorded the highest severity (31.43 %) which did not differ significantly from that of SAMPEA 10 (30.48 %). Dansokoto had the lowest severity (17.14 %) and did not differ significantly from Kanannado, Biu local, IAR-07-1050, SAMPEA 7, SAMPEA 8 and IAR-00-1074.

Table 2: Disease incidence and severity of stem rot on cowpea genotypes inoculated with *Rhizoctonia solani* on the field.

Variety	Disease incidence (%)			Disease severity		
	2WAS	4WAS	6WAS	2WAS	4WAS	6WAS
SAMPEA 10	14.79^{a}	36.90^{ab}	75.41^{a}	9.99^{a}	30.48^{a}	66.19^{a}
IAR-00-1074	6.86^{ab}	24.95^{cd}	48.93^{c}	5.71^{ab}	20.00^{bcd}	26.67^{e}
SAMPEA 12	6.00^{ab}	24.17^{cd}	30.28^{d}	5.71^{ab}	23.81^{bc}	49.52^{b}
IAR-07-1050	6.00^{ab}	26.23^{bcd}	57.91^{bc}	3.56^{ab}	21.90^{bcd}	36.19^{cd}
Biu local	4.19^{ab}	28.97^{bcd}	64.62^{b}	3.18^{ab}	23.81^{bc}	62.86^{a}
SAMPEA 7	0.00^{b}	30.81^{bcd}	50.71^{c}	0.00^{b}	20.00^{bcd}	41.90^{cd}
SAMPEA 8	0.00^{b}	22.43^{d}	51.21^{c}	0.00^{b}	20.95^{bcd}	36.19^{cd}
SAMPEA 9	0.00^{b}	32.58^{a-d}	53.49^{c}	0.00^{b}	19.05^{cd}	43.62^{bc}
SAMPEA 11	0.00^{b}	42.94^{a}	51.74^{c}	0.00^{b}	31.43^{a}	35.24^{d}
Maifarinhanci	0.00^{b}	34.77^{abc}	53.49^{c}	0.00^{b}	24.76^{b}	39.05^{cd}
Kanannado	0.00^{b}	25.29^{cd}	25.29^{d}	0.00^{b}	18.09^{d}	19.05^{f}
Dansokoto	0.00^{b}	21.67^{d}	21.67^{d}	0.00^{b}	17.14^{d}	17.14^{f}
SE±	3.66	3.56	2.99	2.78	1.74	2.40

Means followed by similar superscript in a column are not significantly different at 5 % level of significance ($P \leq 0.05$) using Duncan Multiple Range Test (DMRT)

WAS = Weeks after sowing

At 6 WAS, SAMPEA 10 recorded the highest severity (66.19 %) which did not differ significantly from that of Biu local (62.86 %). Dansokoto recorded the lowest severity (17.14 %) which did not differ significantly from that of Kanannado (19.05 %).

The inoculated cowpea genotypes were separated into different host reaction groups based on the scale described earlier. The glasshouse evaluation result showed that SAMPEA 7, SAMPEA 11 IAR-00-1074, IAR-07-1050 and Biu local were moderately resistant; SAMPEA 8, SAMPEA 9 and SAMPEA 10 were highly susceptible; SAMPEA 12, Maifarinhanci and Kanannado were found to be moderately susceptible while Dansokoto was highly resistant. The field evaluation result however showed that SAMPEA 7, SAMPEA 8, SAMPEA 9, SAMPEA 11, IAR-07-1050 and Maifarinhanci were moderately susceptible; SAMPEA 10, SAMPEA 12 and Biu local were highly susceptible while IAR-00-1074, Kanannado and Dansokoto were moderately resistant (Table 3).

Pod and grain yield loss

The yield loss recorded in the glasshouse significantly varied ($p \leq 0.05$). Biu local recorded the highest pod and grain yield loss. On the field however, IAR-07-1050 had the highest pod and grain yield loss. Dansokoto had the lowest pod yield loss in both glasshouse and field. IAR-00-1074 had the lowest grain yield loss in the glasshouse while SAMPEA 9 had the lowest grain yield loss on the field (Table 4).

Table 3: Host reaction groups of cowpea genotypes evaluated for their reaction to stem rot in the glasshouse and on the field.

Varieties	Host Reaction Group	
	Glasshouse	Field
SAMPEA 7	MR	MS
SAMPEA 8	HS	MS
SAMPEA 9	HS	MS
SAMPEA 10	HS	HS
SAMPEA 11	MR	MS
SAMPEA 12	MS	HS
IAR- 00- 1074	MR	MR
IAR- 07- 1050	MR	MS
Biu local	MR	MS
Maifarinhanci	MS	MS
Kanannado	MS	MR
Dansokoto	HR	MR

MS = moderately susceptible. MR = moderately resistant. HS = highly susceptible.

HR = highly resistant

Table 4: Pod and grain yield loss on cowpea genotypes inoculated with *Rhizoctonia solani* in the glasshouse and on the field.

	Glasshouse		Field	
Variety	Pod yield loss (%)	Grain yield loss (%)	Pod yield loss (%)	Grain yield loss (%)
Dansokoto	3.91^{c}	15.15^{e}	5.9^{6f}	20.16^{de}
IAR-00-1074	8.45^{c}	10.52^{e}	8.81^{def}	17.03^{e}
Kanannado	29.4^{b}	33.88^{cd}	7.50^{ef}	22.24^{cde}
SAMPEA 12	25.34^{b}	36.62^{c}	13.72^{bcd}	12.90^{e}
SAMPEA 8	9.54^{c}	33.26^{cd}	7.38^{ef}	25.40^{cde}
Maifarinhanci	25.03^{b}	35.19^{c}	6.28^{f}	25.10^{cde}
SAMPEA 11	12.11^{c}	14.89^{e}	16.40^{bc}	30.00^{cde}
SAMPEA 7	5.12^{c}	25.50^{d}	12.25^{cde}	48.72^{ab}
SAMPEA 10	22.88^{b}	49.89^{b}	19.49^{b}	36.30^{bcd}
SAMPEA 9	10.83^{c}	25.86^{d}	14.39^{bcd}	17.36^{e}
IAR-07-1050	66.54^{a}	61.15^{a}	47.54^{a}	53.99^{a}
Biu local	70.04^{a}	68.23^{a}	18.12^{b}	39.05^{abc}
SE±	1.55	1.64	1.05	3.16

Means with the same superscript in a column are not significantly different at 5% level of significance ($P \leq 0.05$) using Duncan Multiple Range Test (DMRT)

Table 5 shows the correlation between disease incidence, disease severity, number of pods and grain weight of inoculated cowpea recorded in the glasshouse. Disease incidence was positively and not significantly correlated (0.077) with disease severity, it however correlated negatively and not significant with number of pods (-0.09) and grain weight (- 0.21). Disease severity on the other hand was negatively and significantly correlated with number of pods (-0.76) and grain weight (- 0.57). Number of pods correlated positively and significantly with grain weight (0.63).

Table 5: Correlation between disease incidence, disease severity, number of pods and grain weight of cowpea varieties inoculated with *Rhizoctonia solani* in the glasshouse.

	Disease incidence	Disease severity	Number of pods	Grain weight
Disease incidence	1.00			
Disease severity	0.077	1.00		
Number of pods	- 0.092	- 0.76**	1.00	
Grain weight	- 0.21	- 0.57**	0.63**	1.00

NS = Not significant at 5%** = Significant at 1%

Table 6 shows the correlation between disease incidence, disease severity, number of pods and grain yield recorded on the field. Disease incidence correlated positively and highly significant (0.914) with disease severity, it correlated negatively and significant with number of pods (- 0.42) and grain weight (- 0.40). On the other hand, disease severity correlated negatively and highly significant with number of pods (-0.47) and grain weight (-0.47). Number of pods however correlated positively and highly significant with grain weight (0.67).

Table 6: Correlation between disease incidence, disease severity, number of pods and grain weight of cowpea varieties inoculated with *Rhizoctonia Solani* on the field.

	Disease incidence	Disease severity	Number of pods	Grain weight
Disease incidence	1.00			
Disease severity	0.914**	1.00		
Number of pods	- 0.42*	- 0.47**	1.00	
Grain weight	- 0.40*	- 0.47**	0.67**	1.00

* = significant at 5% ** = significant at 1%

DISCUSSION

It can be deduced from the result presented above that cowpea seedlings even at 2 WAS can be infected by *R. solani* with severity as high as 40 %. There was also a gradual disease progression observed with time which can be attributed to inoculum build-up. The occurrence of disease at 2 WAS which is 1 week after inoculation indicates that inoculation period of *R. solani* is less than a week.

None of the cowpea genotypes tested was immune to the disease even though some varieties showed different level of resistance to the disease. The variability in resistance and susceptibility of the genotypes may be due to differences in their genetic makeup. This agrees with report by Mohammed *et al* that different reactions of cowpea genotypes to bacterial blight, rust and cowpea aphid borne mosaic virus was as a result of genotypic differences (8). Twenty cowpea genotypes tested against rust in Ghana also showed varied resistance response under field condition (17). Higher incidences and severities were recorded on the field than in the glasshouse. The differences are probably attributable to the fact that in the glasshouse experiment, the soil was sterilised and infection was only due to the fungi inoculated; in the field however as the soil was not sterilised, activities of other soil pathogens aggravated the disease making a disease complex that was more severe than single pathogen infection. Study showed that cowpea stem rot is caused by complex of soil fungi which include *S. rolsii*, *R. solani* and *Pythium ultimum* (2). Onuorah (1973) also showed that cowpea stem rot caused by complex of soil borne fungi is more severe than that caused by single fungus (14).

SAMPEA 10, Biu local and IAR-07-1050 are the most susceptible of the 12 cowpea varieties tested. SAMPEA 12, Dansokoto and Kanannado were resistant in both glasshouse and field. With the exception of Biu local, the local varieties recorded relatively lower incidence and severity than the improved varieties, this indicates that the local cowpea varieties tested are more tolerant to stem rot than the improved varieties used in this study. According to IAR released varieties descriptors (4), SAMPEA 10 is resistant to striga/alectra and tolerant to major insect pests. SAMPEA 7 is susceptible to scab, bacterial blight, septoria leaf spot and brown blotch. It is also susceptible to beetles, thrips, pod borer, pod sucking bugs and bruchids. SAMPEA 8 and

SAMPEA 9 are tolerant to major insect pests. SAMPEA 11 is resistant to nematodes and major insect pests. SAMPEA 12 is field tolerant to major insect pests. None of the varieties was earlier evaluated against stem rot. In earlier studies conducted in Sokoto Nigeria, SAMPEA 9 and SAMPEA 11 were found to be highly susceptible to Cercospora leaf spot while SAMPEA 7 and SAMPEA 8 were moderately susceptible to the disease (9)

The result showed that number of pods produced and grain weight significantly decreased with increase in incidence or severity. This shows that cowpea stem rot caused significant yield reduction. This agrees with earlier research that reported negative and significant correlation between cowpea scab severity and grain yield in Samaru Zaria (7, 10). Soonthornpoct *et al.* (2000) reported significant negative correlation between yield of maize and severity of root and basal stem rots incited by *Rhizoctonia solani* in Mississipi, USA (15). Yield of cowpea was also found to be correlated negatively with damping off and brown blotch of cowpea in Zaria (13).

CONCLUSION

From the result of this study it can be concluded that *Rhizoctonia solani* induced stem rot causes significant reduction (17 -53 %) of cowpea yield. Both the local and improved varieties commonly used by farmers in North West Nigeria have different levels of susceptibility/resistance to the disease with more resistance on the local varieties. IAR-00-1074 and Dansokoto found to be resistant in both glasshouse and field are potential breeding materials for resistance of cowpea to stem rot. Grain yield reductions as a result of stem rot infection on cowpea ranges between 17- 53%.

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