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Original Research Article

Evaluation of Selected Cowpea Genotypes for Resistance to Bacterial Blight

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ABSTRACT

Cowpea,
Xanthomonas
axonopodis pv.
vignicola,
Isolates,
Resistance,

Severity

Keywords

Field and screen house studies were conducted with the objective of testing X. axonopodis pv. vignicola isolates on cowpea genotypes for resistance against the bacteria. Isolates were collected from three different cowpea growing zones (Makurdi, Guma and Gboko Local Government Areas) in Benue state. The isolates designated MKD388-1, GUM391 and GBK205-8 respectively were used to produce inocula for screen house and field trials. Hypersensitivity test was conducted on tomato plants while their virulence was compared on 12 cowpea genotypes. The cowpea reactions were rated on a 1-9 scale. Data collected included disease severity and shoot weight. Hypersensitivity test was positive for all and varying reactions were seen among the isolates in their tests for pathogenicity. Disease severity varied significantly (p<0.001) between 1.00 (BOSADP, IT98K-1092-2) and 8.67 (TVx 3236) in pot experiment I at 28 DAI; 1.00 (BOSADP) and 8.33 (Borno Brown) in field experiment I at 42 DAP and similar reactions were seen in BOSADP (1.00) and Borno Brown (8.33) in field experiment II at 28DAI. Three genotypes including BOSADP, IT97K-1092-2 and IT98K-573-2-1 were consistently resistant in both pot and field experiments. Isolates did not differ significantly (p < 0.05) in their virulence means; rating 4.69 (MKD388-1), 4.50 (GUM391) and 4.33 (GBK205-8) respectively. Findings in this study have identified resistance in BOSADP, IT97K-1092-2 and IT98K-573-2-1 in the predominant environment using isolates from targeted areas which can be recommended to farmers in Benue State to enhance protein nutrition and food security.

Introduction

Xanthomonas axonopodis pv. vignicola (Xav) is the causative agent of cowpea bacterial blight (CoBB), a particularly destructive disease of cowpea in Africa (Okechukwu *et al.*, 2010). Symptoms of cowpea CoBB appear as tiny, water-soaked, translucent spots which are more clearly

visible from the abaxial surface of the leaves (Agbicodo *et al.*, 2010). The spots enlarge, coalesce and develop into big necrotic spots, usually with a yellow halo leading to premature leaf drop. The pathogen also invades the stem causing cracking with brown stripes. Pod infection appears as dark

green water-soaked areas, from where the pathogen enters the seeds and causes discolouration and shrivelling (Khisa, 2013).

CoBB is seed-borne (Gena *et al.*, 2009) and the pathogen can be spread by wind –driven rain and insects (Zandjanakon-Tachin *et al.*, 2007), but also crop debris and weeds can play a role as inoculum source (Sikirou and Wydra, 2004).

Among different strategies to control the disease including cultural practices Florini. 1999) (Emechebe and intercropping (Sikirou, 1999; Sikirou and Wydra, 2008), application of chemicals (Kotchoni et al., 2007; Jindal and Thind, 2008), and sowing pathogen-free seeds (Emechebe and Soyinka, 1985); cultivation of resistant cowpea genotypes appears to be a promising strategy with potential to control cowpea bacterial blight (Khatri-Chhetri, 1999; Sikirou, 1999; Emechebe and Lagoke, 2002).

Control strategies based on the use of chemicals are too expensive for low-input farming systems, whilst cultural practices offer mainly long term benefits.

Since 1980, researches mainly conducted by the International Institute of Tropical Agriculture (IITA) and Semi-Arid Food Grain Research and Development (SAFGRAD) led to identification or creation of cultivars exhibiting varying degrees of resistance to bacterial blight (Prakash and Shivashankar, 1982; Sikirou, 1999).

Development of resistant crop varieties requires reliable methods of screening for the trait of interest.

Evaluation under different environmental conditions would also help achieve this goal.

Materials and Methods

Experimental sites

Experiments were conducted in the Molecular Biology Laboratory and Screen House at the University of Agriculture, Makurdi Research Farm respectively $(07^{0}45.985$ 'N. 008⁰37.219'E $07^{0}45.763$ 'N, $008^{0}37.466$ 'E with elevations 112m and 107m) located in the Sudan savannah zone of North-central Nigeria between February 2010 and November 2011.

Sample collection

Infected cowpea leaf samples were first collected in October 2009 from Makurdi zone and secondly in October 2010 from different cowpea growing areas of Benue State in the Southern Guinea savannah agro ecological zone of Nigeria. The collection sites were Makurdi, Guma, and Gboko (Table 1; Figure 1). Infected cowpea leaves and pods which showed early signs of the disease were collected to avoid some saprophytic microorganisms that grow in tissues killed by the primary pathogen (Bobosha, 2003). Samples were collected in clean plastic bags following the sampling method described by Ah-You et al. (2009). Each sample was labelled with all the information including necessary date, location and name of cowpea genotype.

Sources of seed

Cowpea seeds were obtained from the Molecular Biology Laboratory UAM, IITA Ibadan, and IITA Kano. Twelve cowpea genotypes including BOSADP, IT97K-1069-6, TVU 7778, IT98K-1092-2, IT97K-499-35, IT99K-573-1-1, Borno Brown, TVX 3236, IT98K-573-2-1, IT89KD-391,

IT03K-338-1 and IT98K-205-8 were collected and evaluated in the experiments.

Isolation and preservation of X. axonopodis pv. vignicola

Diseased plant tissues were cut in bits and suspended in sterile water for two hours to allow sufficient quantity of bacterial cells to ooze out from the tissue. (Nutrient agar was prepared by dissolving 28g of the agar in 1 litre of distilled water and sterilized by autoclaving at 121°C for 15 minutes). A loopful of the suspension was then streaked on the surface of nutrient agar plates in a zigzag fashion on a quadrant of the plate starting from the circumference (Management of Plant pathogen Collections Australia, 2005). The plates were then inverted and incubated at 27°C for 48-72 hours according to Vauterin et al. (1991). Bacterial colonies from each plate were further sub-cultured repeatedly until pure colonies were obtained. A loopful of each pure culture was streaked on YPSA plates (Yeast extract 5g; Peptone 10g; Sucrose 20g; Agar 12g in 1 litre of distilled water at a pH of 7.4 and autoclaved at 121°C for 15 minutes). The plates were incubated at 28°C for 48 - 72hours. Pure cultures were transferred to YPSA slants incubated at 28⁰ C for 48 - 72hours and preserved at 4° C for further work. In both methods, only round, entire domed and yellowish smooth, bacterial colonies were selected as described by Ah-You et al. (2009).

Hypersensitivity test

Cultures of *Xanthomonas axonopodis* pv. *vignicola* were grown on nutrient agar for 24-48hr. A suspension of each culture plate was made by suspending a loopful of the culture in 1ml of sterilized distilled water and adjusted to an optical density of approximately 0.3-0.4 at 600nm using

spectrophotometer Model no.6131 24475. An aliquot of 2ml of each suspension was injected using a hypodermic syringe into the intercellular spaces of leaves of one-monthold tomato plant. A separate area of the leaf lamina was injected with sterile distilled water as control. The Tomato plants were kept in a screen house at 28-33°C after inoculation until symptoms developed (Durham, 2011).

Layout of pot experiment I: Pathogenicity of X. axonopodis pv. vignicola (Makurdi isolate) on 12 cowpea genotypes planted in pots in Makurdi, Nigeria. experiment was arranged in a completely randomized design (CRD) with three replications. Plastic pots (3347.1 cm²) were filled with sun-dried top loamy soil. Seeds were manually sown at 2.5cm depth on 29th February 2010 at the rate of four seeds per pot, and watered with tap water using watering can. The seedlings were later thinned to two after emergence. Two plants per genotype were left in three sets of which each set consisted of 12 pots giving a total of 72 plants for inoculated and 72 plants for control. Pots were placed on a concrete floor at the University of Agriculture Research One isolate of Xanthomonas axonopodis pv. vignicola (MKD388-1) was used for pathogenicity test.

Experiment II: Pathogenicity of 3 isolates of *X. axonopodis pv. vignicola* (from Makurdi, Guma and Gboko) on cowpea genotypes planted in pots in Makurdi, Nigeria

Three isolates of *Xanthomonas axonopodis pv. vignicola* obtained from three cowpea farming zones of Benue state (Table 1) were used to screen the twelve cowpea genotypes. The layout was also a completely randomized design with three replications. Seeds were sown as stated in section 3.6.1 in

the screen house at the rate of six per pot and later thinned to three per pot after germination i.e. three plants per genotype. This time three plants per genotype were left in three sets of which each set consisted of 12 pots. A total of 108 plants were used for inoculated and 108 plants for control.

Pathogenicity test and disease assessments were done as in section 3.6.3 and 3.6.4 respectively.

Pathogenicity test

Cultures of three bacterial isolates used as inoculum were grown on nutrient glucose agar for 48 hours. After harvesting colonies with sterile distilled water, the concentration was adjusted to an optical density (OD) of 0.3 at 600nm corresponding to 10⁸ colony (CFU/ml) units/ml forming spectrophotometer. Sequential inoculations of the plants were done on the primary and first trifoliate leaves 14 and 21 days after sowing respectively by atomizing a loopful of culture on the lower surface of young expanding leaves after wounding with a brush according to the modified method of Mukesh et al. (2010).

The plants were covered with polyethylene bags and kept in Screen house at 25 - 30°C day and 15-18°C night temperature for 24hr to enhance establishment of infection (Agbicodo *et al.*, 2010). The reaction of plants was observed every week for one month. Noninoculated plants served as control (Plate 1)

Evaluation of disease symptoms in cowpea plants infected with bacterial blight pathogen

Symptom evaluation was carried out by visual inspection of the bacterial-infected plants. The severity was rated on a scale of 1 to 9 according to Marquez *et al.* (2007).

Where,

1= no visible BB symptoms (highly resistant R).

3= isolated chlorotic lesions extending beyond the inoculated area of <25% of inoculated area (resistant R).

5= chlorotic zones around necrotic lesions joined within <50% of the inoculated area (moderately resistant MR).

7= complete chlorosis of inoculated area and chlorotic lesions extending beyond the inoculated area (susceptible S) and

9= severely diseased with large chlorotic lesions also in uninoculated areas (highly susceptible S).

Field experiment

The field assessment was conducted under natural and artificial environments. The trial for each environment was laid out as a randomized complete block design (RCBD) in a sandy-loamy soil at University of Agriculture Research Farm.

Experiment III: Layout under natural infection.

The trials were planted during the period of September – October 2010 with three replications. Each block consisted of 12 ridges divided in two sets. Rows were 2m long and spaced 0.75m apart. Three seeds were sown per hole and later thinned to two; seven days after sowing. On a row there were a total of 12 plants spaced 10cm apart. In this field experiment, trials were scored under natural infection.

Experiment IV: Trial layout under artificial infection

During the planting season April-June 2011, the field was ploughed and harrowed. Trials were arranged with three replications. The field layout had six blocks of 1m each, spaced 1.5m from each other. Every block consisted of 12 rows of plants spaced 75cm apart (Appendix 2). Three seeds were sown per hole, 2cm deep; they were later thinned to two per whole three days after germination leaving 10 plants per row with 10cm spacing between plants (Mukankusi *et al.*, 2010). Plants were inoculated as in the pot experiment 14 days after sowing for the first dose and 21 days after sowing for the second dose. The outer plants on each row were not inoculated to avoid edge effect. Non inoculated plants were used as control.

Field data recording

Disease severity was evaluated at 21, 28, 35 and 42 days after planting (DAP) under natural infection. Subsequently disease assessment under artificial inoculation was carried out using the severity scores in section 3.6.4 at 7, 14, 21 and 28 days after inoculation (DAI). Both fields and screen house data were computed and correlated using the general linear procedure of SAS.

Data analysis

Averages were computed per cowpea genotype on the evaluation day, the highest disease score for either inoculated or non inoculated for each plant was recorded. Cowpeas with mean BB scores of 1 to 3 were considered resistant, and 4 to 6 were intermediate or moderately resistant, and 7 to 9 were considered susceptible. Fodder masses were obtained after assessment period by drying to constant weight under sun to obtain fodder dry weights. The data were used to compute the effect of bacterial blight disease on fodder weight. Disease severity data were computed and analysed using the SAS (version 9.1.3) GLM procedure (SAS Institute, 2001).

Results and Discussion

Hypersensitivity reaction

The inoculated tomato leaves showed positive reaction to all isolates within 3 days (48-72hr) where symptoms of chlorosis to brown necrosis were observed around the inoculated area. This is shown in plate 8. Parts of the leaves inoculated with sterile water remained green.

Pathogenicity reaction

Regarding the pathogenicity test, inoculated leaves of cowpea showed light yellow to brown necrosis around the inoculated/infected area within 7, 14, 21 and 28 days after inoculation/infection, little necrosis was observed in the control. However, only one complete wilting was observed in 4 weeks observation period (Plate 2)

Disease incidence and severity of inoculated plants under pot experiment I and field experiment II

Bacterial blight rating was significantly different (p > 0.05) among the 12 genotypes in pot experiment 1 (Table 2). At 14 days after inoculation, disease scores ranged from 6 to 8 for IT97K -499- 35, TVU 7778, IT89KD-391, Borno Brown, TVx 3236 and IT03K- 338 -1. On the third and fourth assessment periods, disease continued to increase in the susceptible genotypes while BOSADP, IT98K -1092 -2, IT97K- 1069 - 6 and IT98K- 573-2-1 maintained low disease scores which is an indication of resistance. On the contrary, under the field conditions plant stands at 14 days after inoculation were apparently showing increased incidence and severity of bacterial blight. Four cultivars had disease severity scores ranging between 6 and 7.

while two had scores between 5 and 6 and were considered moderately resistant. At the end of the evaluation, all the cultivars that had severity scores between 1 and 4 were considered resistant across the two environments. However, the cultivar, IT98K -205- 8 behaved uniquely because under pot experiment, it appeared moderately resistant but in the field it rated susceptible.

In table 3, the controls placed under disease free conditions showed that cultivars BOSADP, IT98K -1092-2, IT97K -1069- 6, IT98K- 573 - 2 - 1 and IT99K-573- 1-1 consistently showed low severity scores throughout the evaluation period. In both experiments, the susceptible genotypes, IT98K-205-8, IT03K-338-1, TVX 3236 and IT89KD-391 showed systemic expression of bacterial blight as seen on the third and fourth assessment periods across the two environments evaluated. Genotypes TVU 7778 and IT97K-499-35 developed brown leaf spots with limited lesion areas with severity scores ranging between 1 and 2 in the pot 1 control. In field 1 control, the same genotypes TVU 7778 and IT97K-499-35 showed blight spots which enlarged up to 50% of the infected leaf area but no leaves were shed.

Severity and incidence of infection by *X.* axonopodis pv. vignicola under natural and artificial inoculation in the field

The genotypes IT98K -205-8 and TVU 7778 maintained a lower level of susceptibility when allowed to grow under natural infection as compared to the artificial infection. Borno Brown and TVx 3236 had higher bacterial blight scores under artificial infection at 21 and 28DAI compared to the same genotypes under natural infection at 35 and 42DAP. Generally, disease scores ranged between 1 and 5 for cultivars that appeared to be resistant (Table 4). These

cultivars showed good adaptability as well as tolerance to the environmental constraints that occurred including bacterial blight.

Comparative virulence of 3 different *X. axonopodis* pv. *vignicola* isolates on 12 cowpea genotypes

Blight incidence and severity. Table 4 shows the main effect of three bacterial blight isolates on cowpea, the main effect of the isolates, and the interaction effect between genotype/isolate combinations. Analysing the isolate x genotype interactions, disease incidence on the first evaluation day (7DAI) ranged from 3 to 7 with significant differences (p > 0.05) among the inoculated The highest incidence cultivars. in IT89KD-391/MKD recorded 388-1: IT03K-388-1/MKD 388-1 isolate combination and Borno Brown/GUM 391 isolate combination. At 14DAI, disease incidence had reduced giving an implication that some of the cowpea genotypes only displayed a hypersensitive response when challenged with *X*. axonopodis vignicola. However, there was a significant difference (p > 0.05) between the control plants and inoculated cultivars. While the control plants had no incidence of disease, inoculated plants showed incidence ranging from 2 in IT98K-1092-2, BOSADP/GUM 391 isolate combination to 9 in IT03K -338 -1/MKD 388-1 isolate. There were however, no significant differences (p < 0.05) among all the genotype/isolate combinations on the third evaluation period; 21DAI (Table 4).

In Nigeria, cowpeas are grown as rain fed crops and are generally planted during the rainy season when disease development and spread of CoBB are highly favoured (Khisa, 2013). However, the pathogenicity test conducted on the 12 cowpea genotypes and the differences in their reaction to the pathogen indicated differential responses of

cowpea to the bacterial blight pathogen. Our results showed that only limited lesion areas enlarged leading to leaf drop in the most susceptible genotypes. This finding confirms early observations by Gitaitis (1983) about cowpea's defence response mechanism to *X. axonopodis* pv. *vignicola* represented by brown-red discolouration without complete collapse of the tissue.

Cowpea genotype IT98K-205-8 was found moderately resistant in pot experiment but susceptible under field conditions using a single strain of X. axonopodis pv. vignicola and this corroborates the finding of Ajeigbe et al. (2008) who reported that IT98K-205-8 was susceptible under field conditions. A the cowpea genotypes BOSADP,IT98K-1092-2 and IT97K-1096-6 were consistently free of bacterial blight infection and were referred to as resistant thereby confirming early results by Gitaitis (1983) and Ajeigbe et al. (2000) who previously reported IT98K - 1092-2 and IT97K – 1096 – 6 as having high resistance to cowpea bacterial blight. Their data based on bacterial blight scores, placed the resistant varieties in class A (resistant group). Some cowpea cultivars such as IT99 -573 - 1 - 1 and IT98K -573 - 2 - 1 were homogenous in the expression of disease resistance which reflected not only in the sources of the germplasm but also similarity within a population from a single source. Consequently, if population averages were used, cultivars such as BOSADP would have appeared more resistant than the already mentioned cultivars.

Under field disease the evaluation, development was very rapid perhaps because of the uncontrolled conditions which they were allowed to grow. During the first season, disease was allowed to develop under natural infection, disease progression was very slow but became apparent at 28DAP. During the trial, Borno Brown and IT03K - 338 - 1 showed high levels of susceptibility while BOSADP IT98K -1092 - 2. IT97K - 1069 - 6, IT99K 573 - 1 - 1 and IT98K - 573 - 2 - 1 could still be classified as highly resistant at 42DAP. In subsequent field evaluation in season two Borno Brown, IT03K - 338 - 1 showed high levels of susceptibility, this was also evident in the non-inoculated used as control.

In all genotypes, the virulence of the different isolates was similar with no significant difference (p < 0.05). This observation was in contrast to the work done by Shoaga *et al.* (2001) where their result showed significant differences using three different isolates to study seed transmission. In all cases of pot experiments, season two pot experiment gave rise to significantly lower disease incidence compared to season one pot experiment. This is probably because the second pot experiment was conducted in the screen house indicating the absence of confounding effects from the environment.

Table.1 Cowpea bacterial blight samples and areas of collection

Diseased cowpea genotype	Area/zone	Code Name
IT03K-388-1	Makurdi	MKD388-1
IT97K-391	Guma	GUM391
IT98K-205-8	Gboko	GBK205-8

NB: The first three letters are abbreviated zone names The numbers that follow are variety names

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Table.2 Mean scores of bacterial blight (1-9) for cowpea inoculated with *X. axonopodis pv. vignicola* in pot and field experiments

Pot Experiment I (DAI)				-	Field Experiment II (DAI)					
GENOTYPES	7	14	21	28	RT	7	14	21	28	RT
BOSADP	1.00d	2.00g	1.67d	1.00g	R	1.00g	2.00e	1.00h	1.00g	R
Borno Brown	1.00d	7.67ab	7.00bc	7.67abc	S	1.003g	7.33a	9.00a	8.33a	S
IT98K-205-8	2.00c	5.33de	6.67bc	4.33e	MR	5.00ab	6.67a	7.00cd	8.00ab	S
IT97K-1069-6	1.00d	2.00g	2.00d	2.00fg	R	2.33def	3.33cd	4.33g	4.00e	MR
IT03K-338-1	4.00b	8.33a	8.67a	8.00ab	S	5.33a	5.33b	6.00ef	7.00c	MS
1T98K-1092-2	1.67cd	3.33fg	2.67d	1.00g	R	1.33fg	3.00d	1.67h	2.00f	R
TVU 7778	4.00b	6.00cd	7.00bc	8.00ab	S	3.00cd	7.33a	8.00b	8.00ab	S
TVX 3236	3.67b	7.00abc	8.00ab	8.67a	S	4.00bc	6.67a	7.33bc	7.33bc	S
IT89KD-391	5.67a	7.00abc	7.00bc	6.67cd	MS	4.33ab	5.67b	5.33f	6.00d	MS
IT97K-499-35	1.67cd	6.67bcd	7.67ab	7.33cd	MS	1.67efg	3.00cd	6.33de	6.00d	MS
IT98K-573-2-1	2.00c	2.67g	2.33d	2.33f	R	2.00defg	3.00d	4.33g	3.67e	R
IT99K-573-1-1	2.00c	4.33ef	6.00c	6.33d	MS	2.67de	4.00c	1.00h	2.67f	R
F-LSD _{0.05}	0.89	1.40	1.61	1.12		1.05	0.98	0.88	0.68	
CV (%)	21.3	16.0	17.2	12.6		22.0	11.9	10.2	7.5	
p-value	< 0.0001	< 0.0001	< 0.0001	< 0.0001		< 0.0001	< 0.0001	< 0.0001	< 0.0001	

Values are means of three replicates. Means followed by the same letter in each vertical column are not significantly different according to F-LSD at 5%. **RT**: reaction type

Table.3 Mean scores of bacterial blight for cowpea infected with X. axonopodis pv. vignicola under two different field conditions

field experiment inoculated					field under natural infection					
		(DAI)					(DAP)			
Genotype	7	14	21	28	RT	21	28	35	42	RT
BOSADP	1.00g	2.00e	1.00h	1.00g	R	1.00d	1.00f	1.00e	1.00f	R
Borno Brown	1.00g	7.33a	9.00a	8.33a	S	4.33bc	5.33bcd	6.67ab	8.33a	MS
IT98K-205-8	5.00ab	6.67a	7.00cd	8.00	S	1.33d	5.67bc	6.33abc	6.33bc	R
IT97K-1069-6	2.33def	3.33cd	4.33g	4.00e	MR	1.33d	2.33ef	4.00cd	5.67c	R
IT03K-338-1	5.33a	5.33b	6.00ef	7.00c	MS	4.67b	8.67a	8.00a	8.00ab	S
1T98K-1092-2	1.33fg	3.00d	1.67h	2.00f	R	1.33d	3.00def	6.00abc	6.00c	MR
TVU 7778	3.00cd	7.3.3a	8.00b	8.00ab	S	2.33cd	4.00cde	4.33bcd	5.00cd	MR
TVX 3236	4.00bc	6.67a	7.33bc	7.33bc	S	7.33a	7.00ab	6.33abc	6.33bc	S
IT89KD-391	4.33ab	5.67b	5.33f	6.00d	MS	1.67d	5.00bcd	3.33de	3.33de	R
IT97K-499-35	1.76efg	3.67cd	6.33de	6.00d	MS	7.00a	4.67bcde	4.00cd	3.33de	MR
IT98K-573-2-1	2.00defg	3.00d	4.33g	3.67e	R	3.00bcd	4.33cde	2.67de	2.33ef	R
IT99K-573-1-1	2.67de	4.00c	1.00h	2.67f	R	2.33cd	3.33cdef	2.67de	2.00ef	R
F-LSD _{0.05}	1.0491	0.9776	0.8802	0.6754		2.093	2.6103	2.4022	1.9974	
CV (%)	22.1	12.0	10.2	7.5		39.4	34.5	30.8	24.6	
<i>p</i> -value	<0.0001	<0.0001	<0.0001	<0.0001		< 0.0001	0.0009	0.0003	<0.0001	

Data are means of three replicates. Means followed by the same letter in each vertical column are not significantly different according to F-LSD at 5% DAI = Days After Inoculation RT= reaction types: S=susceptible; R= resistant; MS= moderately susceptible; MR= moderately resistant DAP = Days After Planting

Table.4 Comparative virulence of three isolates of Xav on 12 genotypes of cowpea at 21dai

Genotype	G 4 1	•		Genotype	
	Control	1	2	3	Means
IT98K-1092-2	1.00a	1.33	2.67	3.00	2.33e
Borno Brown	1.00a	6.00	5.67	5.33	5.67c
BOSADP	1.00a	1.33	1.33	1.00	1.22f
IT03K-338-1	1.00a	7.67	7.67	3.67	6.22ab
IT89KD-391	1.00a	6.33	6.33	6.00	6.22ab
IT97K-1069-6	1.00a	1.33	1.00	1.67	1.33f
IT97K-499-35	1.00a	5.67	6.67	5.67	6.00abc
IT98K-205-8	1.00a	6.33	5.00	6.33	5.89bc
IT98K-573-2-1	1.00a	2.33	2.00	2.67	2.33e
IT99K-573-1-1	1.00a	2.00	3.00	2.67	2.56e
TVU 7778	1.00a	4.00	4.33	4.67	4.33d
TVX 3236	1.00a	6.67	6.00	6.67	6.44a
Isolate means	1.00	4.25	4.27	4.11	

Values are means of three replicates of disease severity scores associated with makurdi, Guma and Gboko isolates for each genotype. Means followed by the same letter in each vertical column are not significantly different according to F-LSD at 5%. FLSD for Genotype = 0.4838. FLSD for Isolates = 0.2419. FLSD for Genotype x Isolate = 0.1170

Plate.1 Inoculated and non inoculated cowpea in pots



Plate.2 Hypersensitivity of Xav on tomato plant. Plant leaves are twisted out of shape. Arrow point at necrotic patch as a result of BB infection



Figure.1 Pictorial representation of cowpea plants infected with *Xanthomonas axonopodis* pv. *vignicola*



No visible BB **symptoms observed on BOSADP** (highly resistant R).



IT97K-499-35 with an isolated chlorotic lesion extending beyond the inoculated



TVX 3236 showing chlorotic zones around necrotic lesions joined within <50% of the inoculated area (moderately resistant MR).



IT03K-338-1 showing complete chlorosis of inoculated area and chlorotic lesions extending beyond the inoculated area (susceptible S)





IT03K-338-1 severely diseased with large chlorotic lesions

Complete wilting of inoculated TVU 7778 cowpea plant

Based on the individual cowpea bacterial blight scores as well as the DAI, it is evident in this study that the relationship between the different pot and field experiments was highly significant. This may be due to the fact that the experiments were conducted under different seasons and environmental conditions. Time-course and changes in plant performance has been shown to affect the level of resistance to bacterial blight. Cultivars that appeared to have similar levels of resistance at a young stage differed dramatically at an older stage indicating that resistance of seedlings may not reflect resistance in older plants as earlier observed by Okechukwu et al. (2010). This could probably be due to the difficulty in classifying plants late in the season because the hypocotyls are completely covered with lesions making it necessary to score on the basis of the depth rather than percentage of infection (Mukankusi et al., 2010).

However, the ratings in the field were overestimated because they were confounded with the occurrence of other environmental and biotic constraints like insect pests and saprophytic organisms (fungi), thus making field evaluation difficult. From the field results genotypes IT98K-205-8, IT89KD-391 and IT97K-499-

35 produced a moderately resistant reaction with average fodder weight of 1 to 2g while IT99K 573-1-1, IT98K-573-2-1, IT97K-IT98K-1092-2 and BOSADP 1069-6. reacted as resistant and Borno Brown. IT03K-338-1, TVU 7778, and TVX 3236 susceptible reacted as after assessment. It may be tempting to state that similar results will be expected irrespective of the season, X. axonopodis isolates, DAI and DAP evaluations used.

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