African Crop Science Journal, Vol. 26, No. 2, pp. 259 - 275
Printed in Uganda. All rights reserved
ISSN 1021-9730/2018 $4.00
© 2018, African Crop Science Society

African Crop Science Journal by African Crop Science Society is licensed under a Creative Commons Attribution 3.0 Uganda License. Based on a work at www.ajol.info/ and www.bioline.org.br/cs
DOI: http://dx.doi.org/10.4314/acsj.v26i2.8

TRAIT ASSOCIATION AND STABILITY OF VIRUS RESISTANCE AMONG COWPEA GENOTYPES IN UGANDA

E.K. MBEYAGALA[1,2], J. BISIKWA[1], P. TUKAMUHABWA[1] and S.B. MUKASA[1]

[1] School of Agricultural Sciences, Makerere University, P. O. Box 7062, Kampala, Uganda
[2] National Semi-Arid Resources Research Institute (NaSARRI)/NARO, P. O. Box 56, Soroti, Uganda
Corresponding author: kmbeyagala@gmail.com

(*Received 23 October, 2017; accepted 24 May, 2018*)

ABSTRACT

Cowpea (*Vigna unguiculata* L. Walp) grain constitutes an important source of protein for several households in Sub-Saharan Africa. However, widespread occurrence of viral diseases is a serious constraint to productivity of the crop and its nutritional value in terms of grain protein content. This study was carried out to identify cowpea genotypes with the best combination of virus resistance, maturity traits and yield stability. A screening trial for 105 genotypes was established in three locations (Budaka, Tororo and Serere) in Uganda for two consecutive seasons to assess for virus resistance, maturity traits and yield. Season and genotypic effects had a greater contribution to the variation in virus infection among genotypes. Eight genotypes (WC48, NE43, NE15, WC35A, WC39, WC33, WC35C, and WC18) showed low virus infection levels. The three locations formed a single mega-environment with WC51, NE48, and MU17 having the highest mean yield (1,384, 1,191.4 and 1,119.6 kg ha^{-1}), respectively, as well as exhibiting yield stability across locations. Days to first flower, mid-bloom (days to 50% flowering) and days to maturity were positively associated. Virus severity, incidence and AUDPC also had a positive association indicating that indirect selection based on any of these traits is possible. Potential sources of resistance to virus infection exist among the evaluated genotypes. Further screening under high viral pressure is recommended. The high yielding genotypes are recommended for release for cultivation.

Key Words: AUDPC, incidence, severity, *Vigna unguiculata*

RÉSUMÉ

Lesgrains du niébé (*Vigna unguiculata* L. Walp) constituent une source importante de protéine pour un nombre important de ménages en Afrique Sub-Saharienne. Toutefois, la prolifération des maladies virales est une contrainte sérieuse à la productivité de la culture et sa valeur nutritionnelle en termes de la teneur en protéine. Cette étude a été conduite pour identifier les génotypes du niébé avec une meilleure combinaison de résistance au virus, traits de maturité et la stabilité du rendement en grain. Un essai de criblage de 105 génotypes a été établi dans trois localités (Budaka, Tororo et Serere) en Uganda pendant deux saisons consécutives pour évaluer la résistance au virus, traits de maturité et le rendement. Les effets de la saison et du génotypeont contribué beaucoup plus à la variation dans l'infection du virus parmi les génotypes. Huit génotypes (WC48, NE43, NE15, WC35A, WC39, WC33, WC35C, et WC18) ont montré de faibles niveaux d'infection du virus.Les trois localités ont formé un méga-environnement avec WC51, NE48 et MU17 ayant les plus grands rendements moyens (1 384 ; 1 191,4 et1119,6 kg ha-^{1}), respectivement, de même la stabilité du rendement à travers les localités. Le nombre de jours de la première floraison, mid-floraison (nombre de jours de 50% floraison) et le nombre de jours de maturité étaient associés positivement. La sévérité de virus, l'incidence et l'AUDPC ontaussi une association positive indiquant

que la sélection indirecte basée sur n'importe quel de ces traits est possible. Des sources potentielles de résistance à l'infection des virus existent parmi les génotypes évalués. Un criblage sous une forte pression des virus est recommandé. Les génotypes à haut rendement sont recommandés pour délivrance pour production.

Mots Clés: AUDPC, incidence, severité, *Vigna unguiculata*

INTRODUCTION

Cowpea (*Vigna unguiculata* L. Walp) is a key grain legume especially in Sub-Saharan Africa, as an inexpensive source of protein (Timko *et al*., 2007). The grain is highly preferred for its flavour and short cooking time. The crop also is a soil ameliorant through biological nitrogen fixation (Blade *et al*., 1997; Carsky *et al*., 2002). Furthermore, cowpea haulms provide fodder for livestock feeding, especially during the dry season (Blade *et al*., 1997). In addition to the grain, the young leaves/shoots and immature pods are important sources of food and income in Africa (Timko *et al*., 2007).

Given its roles in contributing to food security, income generation and maintenance of the ecosystem, the crop is truly "a multi-functional legume" (Timko and Singh, 2008). In Uganda, 90% of cowpea production takes place in the eastern and northern regions of the country, largely by small holder farmers (Adipala *et al*., 1999). Productivity of the crop is, however low, estimated at less than 500 kg ha^{-1}. This is attributed to a number of biotic and abiotic factors, as well as poor agronomic practices (Edema *et al*., 1997; Adipala *et al*., 1999). Among the biotic constraints, viral diseases which are widespread in the country (Edema *et al*., 1997; Adipala *et al*., 1999; Orawu *et al*., 2005; 2015) contribute greatly to yield reduction. Virus infection of cowpea does not only lead to yield reduction, but also reduces the protein content of the grain (Taiwo and Akinjogunla, 2006).

Breeding for resistance to virus infections is the most cost effective and environmentally friendly approach (Ndiaye *et al*., 1993; Bashir *et al*., 2002). This can be achieved through identification and use of best resistant donors. Selection of genotypes that are good in other aspects on the basis of multiple traits, in addition to disease resistance, is therefore important in crop improvement (Yan and Rajcan, 2002). This study was carried out to identify cowpea genotypes with the best combination of virus resistance, maturity traits and yield stability.

MATERIALS AND METHODS

Study sites and genotypes. This study was carried out during the first and second seasons of 2012 (2012A and 2012B, respectively) at three locations; Serere, Tororo and Budaka districts in eastern Uganda. A total of 105 cowpea genotypes, including local and introduced accessions were evaluated (Table 1). The local genotypes were collected from the northern and eastern regions (N-E), western and central regions (W-C) of the country; while the introductions were obtained from International Institute of Tropical Agriculture (IITA).

An alpha-lattice design was used, with three replicates. Plot size was two rows each of four meters long, with spacing of 60 cm between and 30 cm within rows. Post-flowering pests such as flower thrips (*Megalurothrips sjostedti* Trybom), pod borer (*Maruca vitrata* Fabricius) and pod sucking bugs were controlled by 3-4 sprays using Roket 44 EC (Profenofos 40% and Cypermethrin 4%) starting at the budding stage.

Data collection. Data collection on virus infection commenced three weeks after planting (WAP), and subsequently at 14-days interval until the appearance of the first ripe pods on any of the genotypes. Data were collected on severity of viral symptoms on 10 randomly selected plants in each plot. Disease

TABLE 1. List of cowpea genotypes evaluated in 2012A and 2012B in a varietal study in Uganda

Code	Genotype	Source
G1	EBELAT	NE
G2	IT00K-835-45	IITA
G3	IT03K-124	IITA
G4	IT04K-219-2	IITA
G5	IT04K-221-1	IITA
G6	IT04K-227-4	IITA
G7	IT06K-121	IITA
G8	IT06K-123-1	IITA
G9	IT06K-124	IITA
G10	IT06K-147-1	IITA
G11	IT06K-154-1	IITA
G12	IT06K-281-1	IITA
G13	IT06K-91-11-1	IITA
G14	IT07K-187-24	IITA
G15	IT07K-188-49	IITA
G16	IT07K-211-1-8	IITA
G17	IT07K-243-1-5	IITA
G18	IT07K-292-10	IITA
G19	IT07K-299-4	IITA
G20	IT07K-300-12	IITA
G21	IT89KD-288	IITA
G22	IT97K-499-35	IITA
G23	IT98K-503-1	IITA
G24	MU09B	MAK
G25	MU15	MAK
G26	MU17	MAK
G27	MU19	MAK
G28	MU20	MAK
G29	MU20B	MAK
G30	MU9	MAK
G31	NE13	NE
G32	NE15	NE
G33	NE17	NE
G34	NE18	NE
G35	NE19	NE
G36	NE23	NE
G37	NE30	NE
G38	NE31	NE
G39	NE32	NE
G40	NE36	NE
G41	NE37	NE
G42	NE39	NE
G43	NE4	NE
G44	NE40	NE
G45	NE41	NE
G46	NE42	NE
G47	NE43	NE
G48	NE44	NE
G49	NE45	NE
G50	NE46	NE
G51	NE48	NE
G52	NE49	NE
G53	NE5	NE
G54	NE50	NE
G55	NE51	NE
G56	NE53	NE
G57	NE55	NE
G58	NE6	NE
G59	NE67	NE
G60	NE70	NE
G61	NE71	NE
G62	SECOW2W	E
G63	WC1	WC
G64	WC10	WC
G65	WC11	WC
G66	WC12	WC
G67	WC13	WC
G68	WC15	WC
G69	WC16	WC
G70	WC17	WC
G71	WC18	WC
G72	WC2	WC
G73	WC21	WC
G74	WC26	WC
G75	WC27	WC
G76	WC29	WC
G77	WC30	WC
G78	WC32	WC
G79	WC33	WC
G80	WC35A	WC
G81	WC35B	WC
G82	WC35C	WC
G83	WC36	WC
G84	WC39	WC
G85	WC4	WC
G86	WC41	WC
G87	WC42	WC
G88	WC44	WC
G89	WC48	WC
G90	WC5	WC
G91	WC51	WC
G92	WC52	WC
G93	WC53	WC
G94	WC55	WC
G95	WC6	WC
G96	WC62	WC
G97	WC63	WC
G98	WC64	WC
G99	WC65	WC
G100	WC66	WC
G101	WC67	WC
G102	WC67A	WC
G103	WC68	WC
G104	WC69	WC
G105	WC7	WC

Key: N-E = North-eastern Uganda; E = Eastern Uganda; W-C = Western and Central Uganda; MAK = Makerere collection; IITA = International Institute of Tropical Agriculture

severity was based on visual estimation of the diseased plants, as manifested by the different symptoms, on a scale of 1-5 [1 = no symptoms on all leaves, 2 = slight symptoms (1 to 25% of the leaves infected), 3 = moderate symptoms (26 to 50% leaves infected), 4 = prominent symptoms with stunting (51 to 75% of leaves infected), 5 = highly severe symptoms with stunting (> 75% of leaves infected)] (Gumedzoe *et al*., 1997). Virus incidence data was recorded as percentage of symptomatic/ diseased plants in each plot as:

$$\frac{\text{Number of diseased plants}}{\text{Total number of plants}} \times 100$$

Data on agronomic traits such as days to first flowering, days to 50% flowering (mid-bloom) and days to physiological maturity were collected as the number of days between planting date and date of appearance of first flower, date at 50% flowering and date at physiological maturity, respectively. At maturity, pods from all plants in each plot were harvested, sun-dried, and threshed, and weighed to obtain plot seed yield. Plot seed yield was used to determine yield per hectare by extrapolation.

Data analyses. Virus severity data were used to compute area under disease progress curve (AUDPC), as described by Campbell and Madden (1990), *viz*:

$$\text{AUDPC} = \sum_{i=1}^{n-1} \left(\frac{Y_i + Y_{i+1}}{2}\right)(t_{i+1} - t_i)$$

Where:

n = number of successive readings, Y*i* = severity at time *i*, t_i = number of days after the first observation on assessment date *i*.

Disease data (AUDPC, incidence, as well as agronomic data) were subjected to analysis of variance (ANOVA) across locations, using "agricolae" package (Felipe de Mendiburu, 2017). AUDPC and yield values were also analysed using the genotype, and genotype by environment (GGE) biplots, to understand the effects of genotype (G) and genotype by environment interactions (Yan *et al*., 2000; Yan, 2005).

The associations between viral infection and other traits was assessed using genotype by trait (GT) biplots (Yan and Rajcan, 2002). GGE and GT biplot analyses were carried out using "GGEBiplotGUI" package (Frutos *et al*., 2014). All analyses were implemented using R Software Version 3.4.1 (R Core Team, 2017). GGE biplot analysis was based on model/ equation 7 (Yan, 2005) expressed as;

$$P_{ij} = Y_{ij} - \mu - \beta_j = \alpha_i + \phi_{ij} \quad \text{..... (1)}$$

Where:

i is the value of genotype in environment *j*, Y_{ij} is the genotype by environment two way table, μ is the grand mean, α_i is the genotype (row) main effect, β_j is the environment (column) main effect, φ_{ij} is the specific genotype by environment interaction, P_{ij} is the matrix that is subjected to singular value decomposition (SVD).

For GT biplot analysis, model 2 (Yan and Rajcan, 2002) was used expressed as:

$$\frac{T_{ij} - \overline{T}_j}{S_j} = \lambda_1 \delta_{i1} T_{j1} + \lambda_2 \delta_{i2} T_{j2} + \varepsilon_{ij} \quad \text{..... (2)}$$

Where:

T_{ij} is the average value of genotype *i* for trait *j*, $\overline{T}_j$ is the average value of trait *j* over all genotypes, S_j is the standard deviation of trait *j* among the genotype averages; λ_1 and λ_2 are the singular values for the first and second principal components, PC1 and PC2 respectively; δ_{i1} and δ_{i2} are the PC1 and PC2 scores, respectively, for genotype *i*; T_{j1} and T_{j2} are the PC1 and PC2 scores, respectively, for

trait *j*; and ε_{ij} is the residual of the model associated with the genotype *i* in trait *j*.

RESULTS

Area under disease progress curve. Genotypes, locations and seasons significantly (P<0.001) influenced the virus area under disease progress curve (Table 2). Interactions between genotypes and locations, genotype and season, location and season, as well as genotype by site by season also significantly influenced disease progress (P<0.001 and P<0.01, respectively) (Table 2). However, the greatest variation in disease progress was due to the effect of season (63.6%); followed by genotypic effects at 12.5%.

Genotype and genotype by environment biplot analysis of AUDPC data explained 93.06% of the variation in the data (Fig. 1). The first principal component (PC1/Axis1) accounted for 81.15%; while the second principal component (PC2/Axis2) accounted for 11.19%. The "which-won-where" feature of the GGEBiplotGUI package generated a biplot (Fig. 1) with seven sectors, with the most responsive (most susceptible) genotypes for each sector located at the respective vertex. The vertex genotypes were the most responsive, since they had the longest distance from the biplot origin.

From Figure 1, the three locations (Serere, Tororo and Budaka) formed a single mega-environment with regard to AUDPC. In this mega-environment, the most responsive (most susceptible) genotypes were G50 (NE46), G13 (IT06K-91-11-1), G15 (IT07K-188-49), G14 (IT07K-187-24), G11 (IT06K-154-1), G6 (IT04K-227-4), G19 (IT07K-299-4, and G22 (IT97K-499-35); while the most resistant were G89 (WC48), G47 (NE43), G79 (WC33) and G71 (WC18).

It was also observed that genotypes G89, G32, G47, G71, G79, G82, G80, G84, and G63 had the lowest AUDPC values (Fig. 1). Ranking of cowpea genotypes on the basis of AUDPC, using a concentric circle biplot (Blanche *et al.*, 2007) placed the most susceptible genotypes (with high AUDPC values) at or close to the center of concentric circles (Fig. 2), along the average tester axis (ATA). Accordingly, the most susceptible genotypes were G50, G13, G15, G7, G28, G10, G8, G18, G14, G20, G21, G11, G6, G3 and G9. On the other hand, genotypes further to the right on the ATA (opposite direction of the single arrow) and further below the stability axis had the lowest AUDPC values and thus

TABLE 2. Area under disease progress curve (AUDPC) across locations and seasons in a study of cowpea genotypes in eastern Uganda

Source of variation	Df	AUDPC	
		Mean square	Variation (%)
Blocks	9	1214.99***	0.88
Replicates	2	251.72ns	0.04
Genotypes	104	1488.17***	12.47
Sites	2	18452.16***	2.97
Seasons	1	788616.43***	63.56
Genotype x site	208	217.25***	3.64
Genotype x season	104	350.18***	2.94
Site x season	2	7525.95***	1.21
Genotype x site x season	208	134.161**	2.25
Residuals	1228	101.36	10.03

***, **, * = significant at P<0.001, 0.01 and 0.05, respectively; and ns = non-significant at 0.05

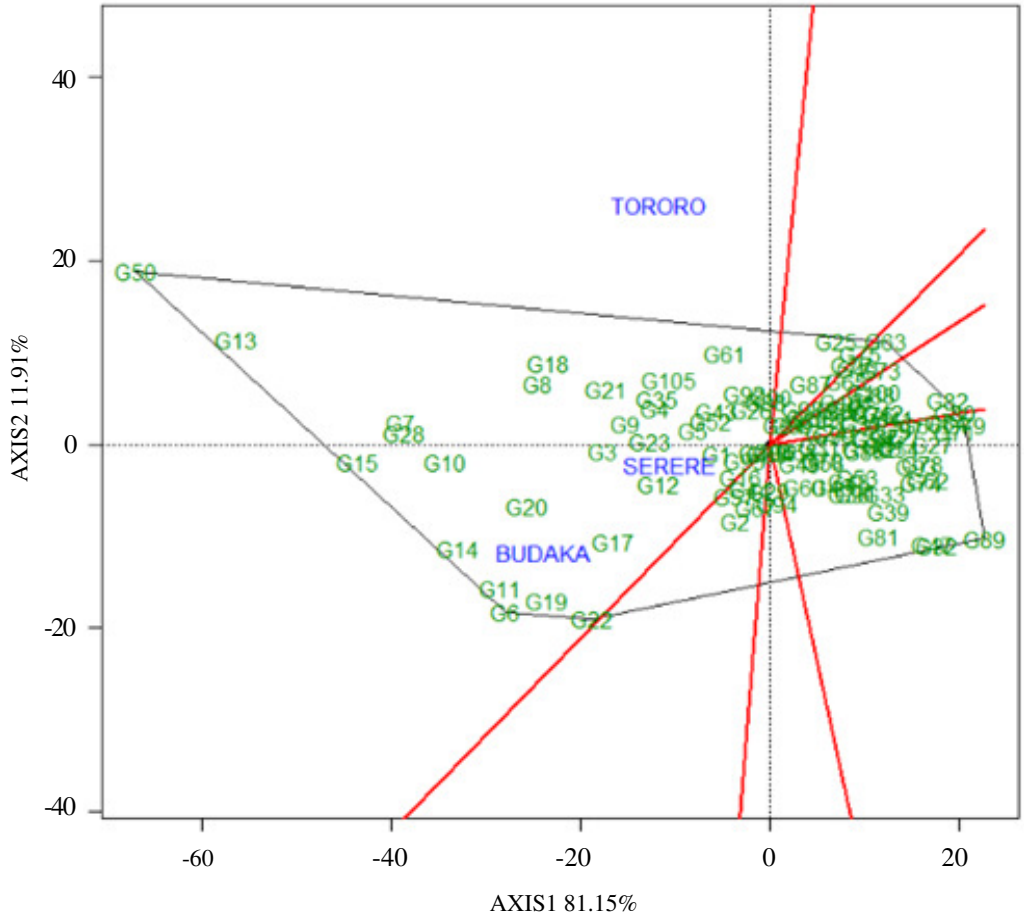

Figure 1. Mega-environment and most responsive genotypes based on AUDPC among cowpea genotypes across three locations in eastern Uganda using a genotype focused biplot.

the least susceptible. These genotypes included G89, G47, G32, G80, G84, G79, G82 and G71.

Virus incidence. The effects of blocks, genotypes, location and season significantly ($P<0.001$) influenced virus incidence (Table 3). The interactions between genotype and location ($P<0.05$), genotype by season ($P<0.001$) and location by season ($P<0.001$) also significantly influenced the level of virus incidence. However, genotype by site by season interactions had no significant influence on incidence. Season effects contributed to the greatest variation of virus incidence at 28.5% followed genotypic effects at 24.5% (Table 3).

When incidence data were subjected to GGE biplot analysis ("which, won, where" feature), the resulting biplot explained up to 93.96% of the variation in the data (Fig. 3); PC1 explained 86.58%, while PC2 explained 7.38% of the variation in the data.

The three locations were grouped into two sectors/mega-environments. The first sector comprised of Serere and Tororo; while the second sector comprised of only Budaka. Genotypes G20 and G30 had the highest virus incidence in the first mega-environment; while genotypes G50, G13, G7, G10, G18, G11 had

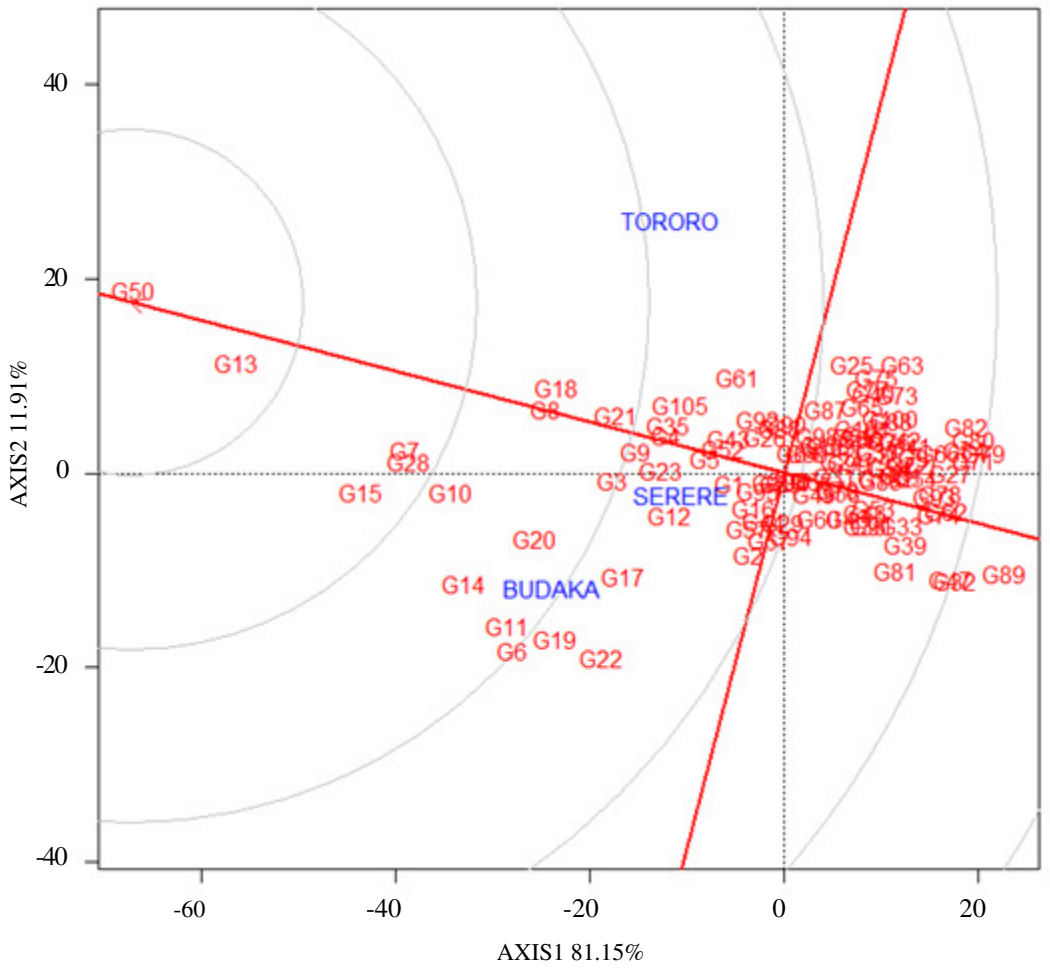

Figure 2. Ranking of genotypes in relation to the "most susceptible" to viral infection based on AUDPC values using a genotype focused biplot in a cowpea varietal study in Uganda.

the highest disease incidence in the second mega-environment (Fig. 3). On the other hand, genotypes G80, G71, G79, G82, G72, G84, G27 and G89 had the lowest virus incidence levels.

When genotypes were ranked using concentric circle biplot (Fig. 4), genotypes further to the left in the direction of the arrow on the ATA had high virus incidence level; while those to the right (opposite of the arrow) had low incidence levels (Fig. 4). Genotype G13 had the highest virus incidence (at the center of the smallest concentric circle); followed by G50, G20, G28 and G7. On the other hand, G89, G82, G71, G79 and G74 further to the right on the ATA and below the stability axis had the lowest incidence.

Yield and other agronomic traits. Genotypes, location and site by season interactions significantly ($P<0.001$) contributed to the variation in yield, days to first flower and days to 50% flowering as revealed by high percentage of explained variation (Table 4). Block and replicate effects also significantly influenced yield, but their contributions to explained variation were minimal. For the case of days to 75% maturity,

TABLE 3. Variation for virus incidence across locations and seasons in a study of cowpea genotypes in eastern Uganda

Source of variation	Df	Incidence	
		Mean square	Variation (%)
Blocks	9	551.13***	0.46
Replicates	2	352.75ns	0.07
Genotypes	104	2557.32***	24.5
Sites	2	40743.99***	7.51
Seasons	1	309086.11***	28.48
Genotype x site	208	201.67*	3.86
Genotype x season	104	413.83***	3.97
Site x season	2	46850.17***	8.63
Genotype x site x season	208	183.24ns	3.51
Residuals	1228	168.01	19.01

***, **, * = significant at P<0.001, 0.01 and 0.05, respectively and ns = non-significant at 0.05

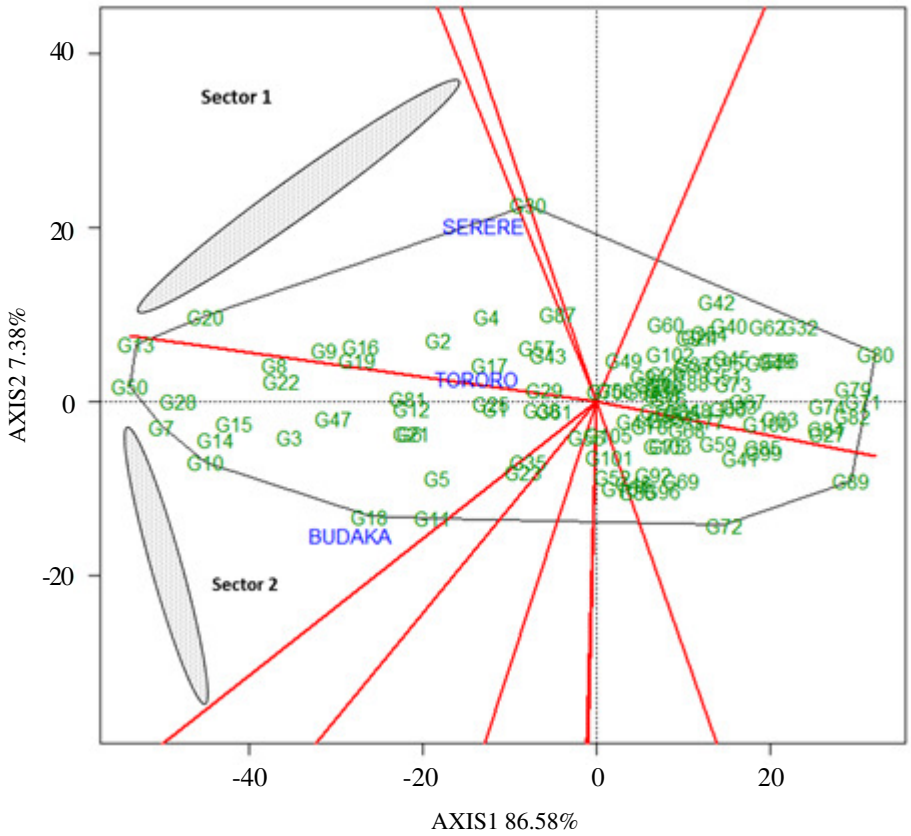

Figure 3. Mega-environments and most responsive genotypes for virus incidence in cowpea genotypes across locations in eastern Uganda using a genotype focused biplot.

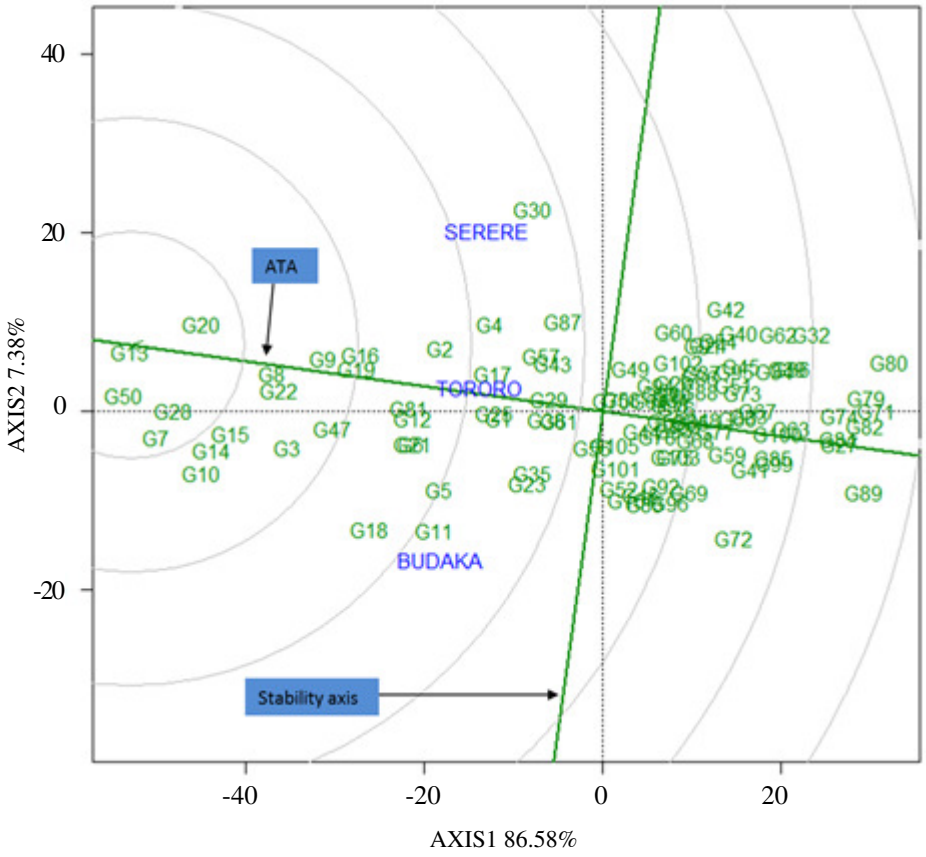

Figure 4. Ranking of cowpea genotypes based on virus incidence using a genotype focused concentric circle biplot in a cowpea varietal study in eastern Uganda.

season, genotype, genotype by site interactions, site and site by season and interactions had the greatest significant contribution to explained variation (of 9.15, 9.29, 9.33, 17.17 and 31.52%) ($P<0.001$).

Analysis of yield data using GGE biplot revealed that the axis1 (PC1) explained 67.84%; while axis2 (PC2) explained 17.73% of the total G and GE variation in grain yield (Fig. 5). Therefore, the first two axes explained 85.57% of the total variation. The three study locations formed a single mega-environment, with genotypes; G91 (WC51), G41 (NE37), G51 (NE48) and G54 (NE50) genotypes as winning genotypes. The average yield, for G91 (WC51), G41 (NE37), G51 (NE48) and G54 (NE50) were; 1384.7, 1156.5, 1191.4 and 976.2 kg ha^{-1}, respectively.

To identify desirable genotypes (both high yielding and stable across locations), a concentric circle biplot (Fig. 6) was used to rank genotypes in relation to the "ideal genotype". An "ideal" genotype (center of the innermost concentric ring/circle) is a point on the AEA ("absolutely stable") in the positive direction and has a vector length equal to the longest vectors of the genotypes on the positive side of ATA ("highest mean performance") (Yan and Tinker, 2006). Thus, G91, G51, G26, G78, G55, G41, G72, G87, G32, G93, G62 and G54 were closer to the "ideal genotype" and, therefore, more desirable than others. On the other hand, the poorest genotypes were G22, G29, G20, G95, G65, G19 and G90.

TABLE 4. Mean squares for yield and other agronomic traits for cowpea genotypes evaluated in three locations.

Source of variation	Df	Yield		First flower		Mid-bloom		Maturity	
		Mean square	Percent	Mean square	Percent	Mean square	Percent	Mean square	Percent
Blocks	9	451420.92***	0.57	52.46***	1.03	61.487***	1.03	40.15***	0.41
Rep	2	410516.43*	0.12	30.85*	0.14	74.96***	0.28	10.71ns	0.02
Genotype	104	733000.37***	10.68	83.10***	18.91	91.84***	17.73	78.32***	9.29
Site	2	53031087.44***	14.86	3637.20***	15.92	4846.16***	17.99	7530.83***	17.17
Season	1	15413989.41***	2.16	1590.32***	3.48	482.30***	0.89	8025.73***	9.15
Genotype x site	208	224969.52***	6.56	14.36***	6.54	20.69***	7.99	39.36***	9.33
Genotype x season	104	278436.75***	4.06	12.05***	2.74	15.35***	2.96	31.66***	3.75
Site x season	2	106948584.78***	29.97	6463.67***	28.29	8492.84***	31.54	13821.15***	31.52
Genotype x site x season	208	286842.12***	8.36	10.23***	4.66	11.14***	4.31	20.23***	4.77
Residuals	1227	131916.87	22.68	6.88	18.29	6.77	15.27	10.62	14.57
Range		8.33-3000		34-78		39-73		67-101	

***, **, * = significant at P<0.001, 0.01 and 0.05, respectively; and ns = non-significant at 0.05

Relationships between cowpea traits. Across all genotypes evaluated, virus incidence, severity and area under disease progress had a positive association as indicated by the acute angles between their vectors (Fig. 7). Days to first flowering, days to 50% flowering (mid-bloom) and days to 75% maturity also had acute angles between their vectors and, therefore, were positively correlated. However, there was a strong negative association between AUDPC and yield, average severity and yield, and incidence and yield as indicated by the large obtuse angles between their angles. It was observed that there was a near zero correlation between yield and days to first flowering, between yield and mid-bloom and between yield and maturity as indicated by the near perpendicular vectors (Fig. 7). Also, the three disease variables (AUDPC, severity and incidence) had a near zero correlation with all the maturity related variables (first flowering, mid-bloom and maturity) as indicated by perpendicular vectors.

Genotypes G50, G13, G7, G21 and G17 constituted a group of genotypes with similar trait profiles of high area under disease progress, high incidence and high severity values often with yield below the average mean. On the other hand, G89, G41, G84, G72, G93, and G78 were associated with low to medium virus infection levels, higher yields and similar maturity periods.

Not all genotypes that were resistant to virus infection were high yielding (Table 5). For example, G50 (NE46) which had the highest AUDPC, severity and incidence values yielded higher (929 kg ha^{-1}) than the overall mean (670 kg ha^{-1}) across the three locations. The yield of G50 was also higher than that of G89 (WC48) which was the most resistant genotype.

DISCUSSION

Area under disease progress and virus incidence. Seasonal differences had the greatest effect on AUDPC and incidence (Table

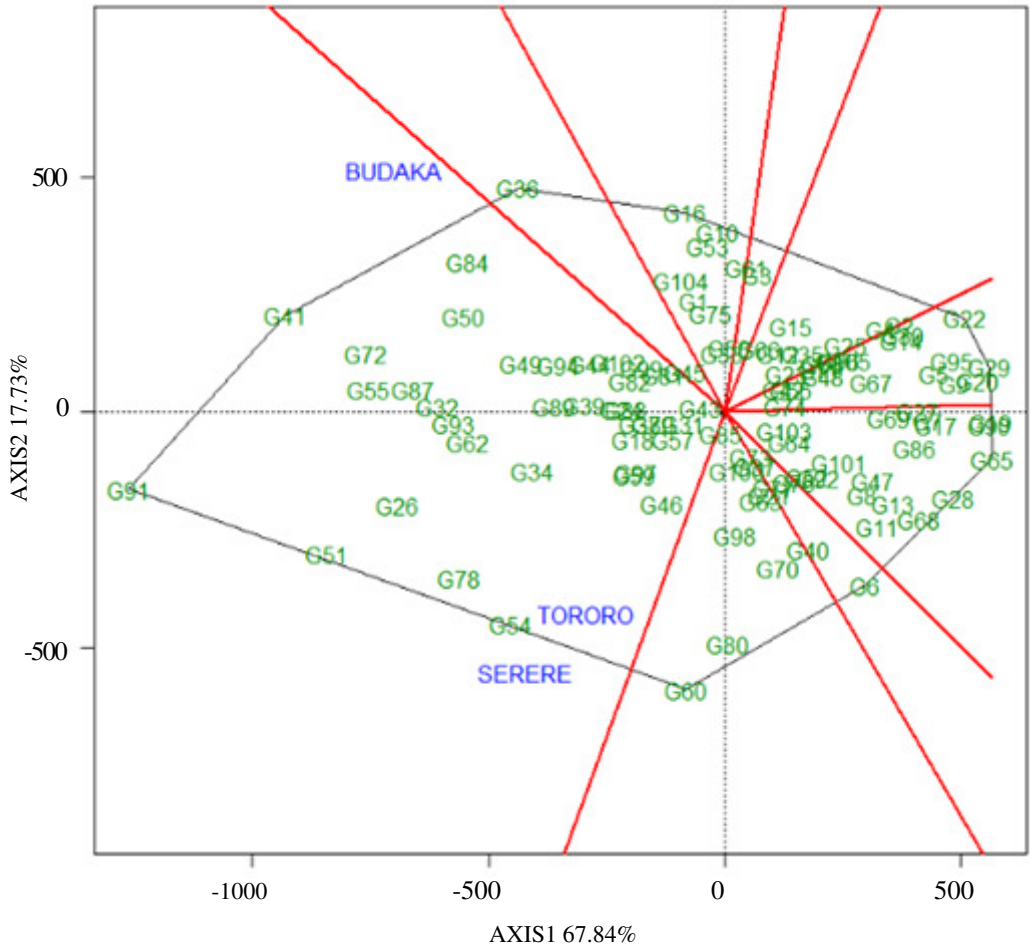

Figure 5. Biplot view of mega-environments and winning genotypes for grain yield using a genotype focused biplot in a cowpea study in eastern Uganda.

2 and Table 3) with lower virus incidence and AUDPC in the first season compared to the second season. However, this was contrary to the observations of Edema *et al*. (1997) and Adipala *et al*. (1999), who reported higher severities and incidences of viral diseases in the first season (wet season) than in the second dry season. Dry weather conditions are associated higher virus vector infestations due to high vector populations and greater mobility than the wet seasons (Edema *et al*., 1997; Taiwo *et al*., 2006; Kone *et al*., 2017a,b). Edema *et al*. (1997) and Adipala *et al*. (1999) attributed the higher viral incidence in the first (wet) season to short dry spells.

Genotypic effects were the second contributor to the total explained variation after season effects, indicating the presence of genetic variation among the genotypes. It is, therefore, possible to obtain sources of resistance among the existing pool of genotypes. Previous studies by Edema *et al*. (1997), Goenaga *et al*. (2008), Aliyu and Balogun (2011), also reported the influence of cowpea variety differences on viral disease severity and incidence.

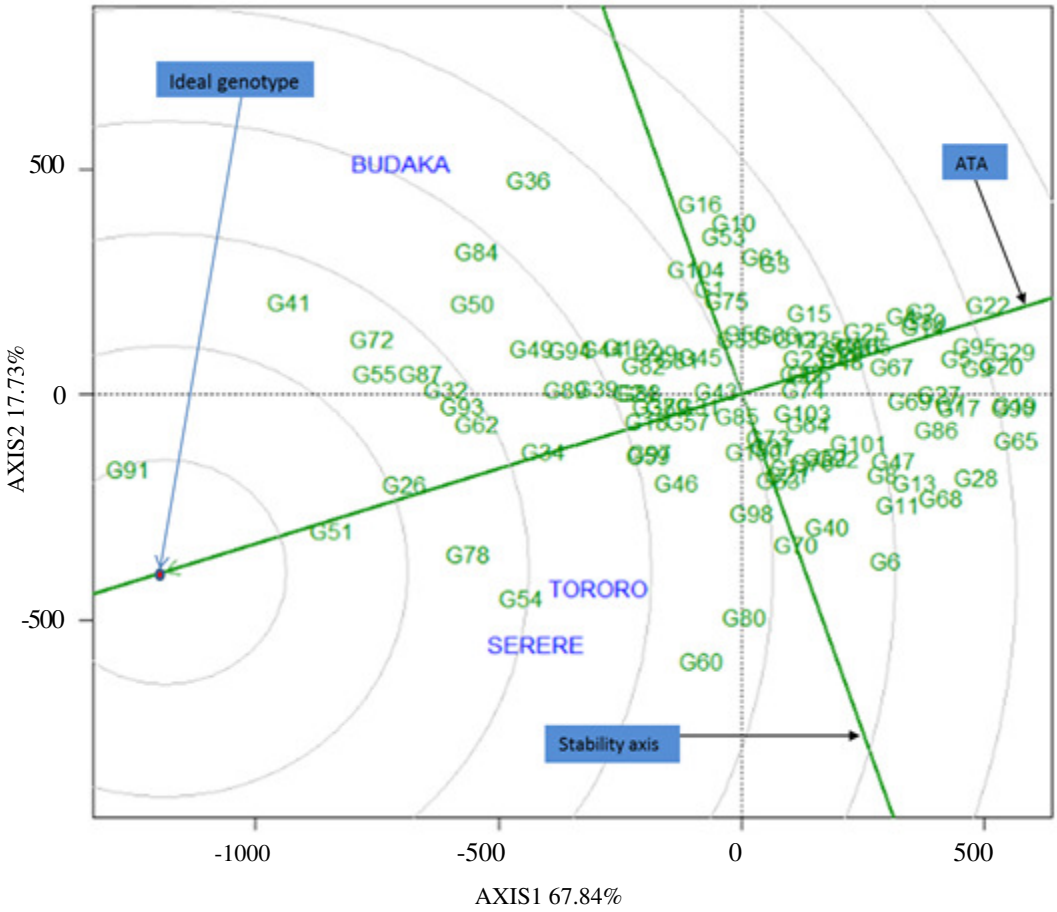

Figure 6. Ranking of cowpea genotypes relative to the ideal genotype for grain yield using a genotype focused concentric circle biplot in a varietal study in eastern Uganda.

In this study, it was possible to graphically identify susceptible or responsive genotypes (G50, G13, G15, G14, G11, G6 and G22), as well as resistant genotypes (G89, G32, G47, G79, G80, G82, G71, G84, and G62) (Figs. 1 - 4). Studies by Yan and Rajcan (2002), Yan and Tinker (2006) and Blanche *et al*. (2007) also reported on discriminative power of GGE biplots. Though none of the genotypes was completely immune to virus infection as previously reported by Mbeyagala *et al*. (2014), the identified resistant genotypes are potential sources of resistance for germplasm improvement, as well as cultivation if preferred by farmers.

Yield and other traits. Genotype, locations and location by season interactions had the greatest contribution to variation in yield and maturity indices (Table 3). This observed variability allows for selection of suitable genotypes for grain yield and maturity periods as previously reported (Hawkes, 1991).

Based on the median values for 75% maturity, most of the cowpea genotypes in this pool are medium maturing. The range of yield values indicated that both low to potentially high yielding genotypes exist in this collection. These findings collaborate those reported by Agbahoungba *et al*. (2017); thus it is possible to select genotypes with high yield potential from the assembled germplasm pool.

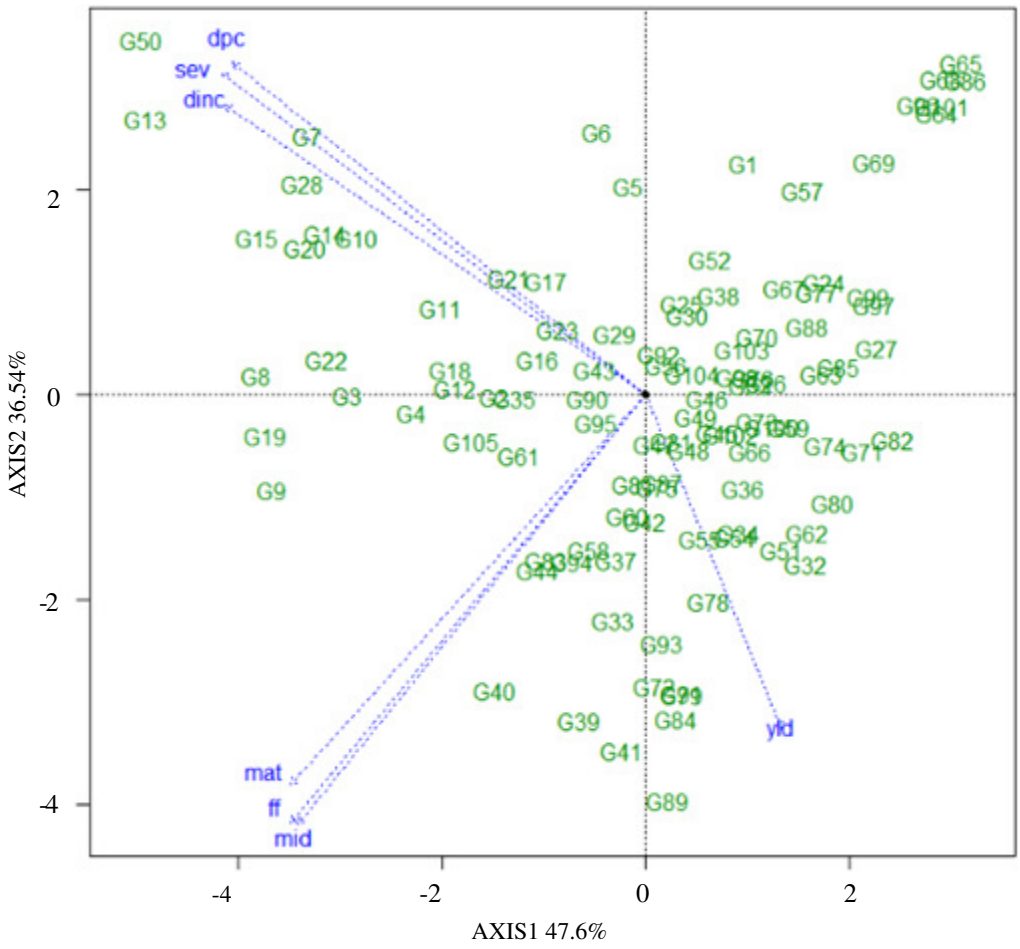

Figure 7. Genotype by trait biplot of 105 cowpea genotypes for seven traits across three locations (Budaka, Tororo and Serere) in eastern Uganda. yld = yield (kg ha^{-1}), mid = mid-bloom, ff = days to appearnce of first flower, mat = days to 75% maturity, dinc = incidence, sev = average severity, dpc = area under disease progress.

The three locations formed a single mega-environment (Fig. 5), with G91, G41, G51, G26 and G54 as the high yielding and stable genotypes (Fig. 6). Ceccarelli (2012) suggested that good genotypes should have the best combination of high yield and high stability. Partitioning of test locations into mega-environments and deploying different genotypes for different mega-environments, is the best way to exploit positive genotype x environment interactions, while avoiding negative ones (Yan and Tinker, 2006). However, data from multiple seasons are required to verify the repeatability of the observed mega-environment pattern (Yan and Tinker, 2005, 2006).

Associations between traits. The genotype by trait biplot (GT) effectively revealed the interrelationships among cowpea traits (Fig. 7) and, thus helped to identify traits that were positively or negatively associated. For instance, there was a positive association between severity, incidence and AUDPC, as well as positive association between days to first flowering, mid-bloom and days to 75% maturity (Fig. 7). From the GT biplot, it was observed that some of the traits measured were

TABLE 5. Means of AUDPC and other traits for 10 most resistant (top) and 10 most susceptible (bottom) cowpea genotypes at the three locations

Code	Genotype	AUDPC	Yield	Severity	Incidence	Number of days to first flower	Number of days to 50% flowering	Number of days to 75% maturity
G89	WC48	39.77	865.20	1.50	23.64	52.18	55.42	86.42
G47	NE43	42.56	538.66	1.58	58.41	48.94	52.22	83.33
G79	WC33	42.71	744.68	1.62	24.30	51.92	55.67	83.36
G71	WC18	42.86	558.70	1.55	23.76	48.12	51.41	80.64
G32	NE15	43.26	1004.54	1.66	28.49	48.39	52.20	82.67
G62	SECOW2W	44.00	961.67	1.65	30.54	48.47	51.41	82.54
G80	WC35A	44.26	764.12	1.65	23.40	48.50	52.11	81.09
G27	MU19	44.28	455.09	1.67	25.80	46.72	50.00	80.39
G74	WC26	44.28	622.82	1.69	25.76	46.94	51.28	82.40
G84	WC39	44.32	916.76	1.60	25.85	50.61	54.91	85.66
G50	NE46	94.72	929.17	3.57	71.86	47.61	52.11	83.17
G13	IT06K-91-11-1	86.88	502.55	3.11	71.92	49.19	53.11	84.33
G15	IT07K-188-49	77.43	558.06	2.75	64.88	50.37	53.60	83.50
G7	IT06K-121	75.31	444.01	2.74	69.60	48.72	51.76	83.10
G28	MU20	73.38	426.62	2.67	68.60	48.86	53.61	82.75
G14	IT07K-187-24	70.60	436.76	2.56	65.82	49.08	53.72	83.36
G10	IT06K-147-1	69.87	608.06	2.58	66.30	49.11	53.11	82.74
G8	IT06K-123-1	69.27	531.76	2.54	62.49	51.28	54.26	86.13
G20	IT07K-300-12	66.51	376.85	2.61	67.66	48.52	53.36	85.37
G6	IT04K-227-4	66.30	581.94	2.35	53.01	46.56	48.75	81.13

redundant; for instance maturity traits (days to first flower, mid-bloom and days to maturity) and virus infection traits (severity, incidence and AUDPC). This indicates that any of these traits can be used to indirectly select for maturity and virus resistance. The use of GT biplots to describe interrelationships among traits has been reported previously for other crops such as soybean (Yan and Rajcan, 2002), barley (Yan and Tinker, 2005), wheat (Yan and Tinker, 2006) and forage sorghum (Aruna *et al*., 2016).

CONCLUSION

Our results showed that season and genotypic effects contributed greatly to the variation in response to virus infection in cowpea. Genotypes G89, G47, G32, G80, G84, G79, G82, and G71 showed low virus infection levels; thus could be used in cowpea improvement for resistance. These genotypes need to be subjected to enhanced virus levels, through artificial inoculation, to ascertain their reaction.

Based on yield data, the three locations formed a single mega-environment with G91 (1,384 kg ha^{-1}), G51 (1,191.4 kg ha^{-1}), and G26 (1,119.6 kg ha^{-1}) exhibiting stability and high yield. However, multi-season data are needed to confirm this pattern of mega-environment delineation. The high yielding genotypes can be recommended for farmer cultivation after evaluation for acceptability. Positive correlations exist among the three maturity indices (days to first flowering, mid-bloom and days to 75% maturity) as well as among the three virus reaction indices (severity, incidence and AUDPC) suggesting that indirect genotype selection based on any of those traits in each category is possible.

ACKNOWLEDGEMENT

This study was supported with funding from the McKnight Foundation, through the Collaborative Crop Research Program (CCRP). We are grateful to the partial funding provided by Carnegie Cooperation of New York (Grant # RU/2016 /Carnegie/DFS/001) through the Regional Universities Forum for Capacity Building in Agriculture (RUFORUM).

REFERENCES

Adipala, E., Omongo, C.A., Sabiti, A., Obuo, J.E., Edema, R., Bua, B., Atyanga, A., Nsubuga, E.N. and Ogenga-Latigo, M.W. 1999. Pests and diseases on cowpea in Uganda: Experiences from a diagnostic survey. *African Crop Science Journal* 7(4):465-478.

Agbahoungba, S., Karungi, J., Odong, T.L., Badji, A., Sadik, K. and Rubaihayo, P. R. 2017. Stability and extent of resistance of cowpea lines to flower bud thrips in Uganda. *African Crop Science Journal* 25(1): 1-24. https://doi.org/10.4314/acsj.v25i1.1

Aliyu, T.H. and Balogun, O.S. 2011. Effects of variety and planting density on the incidence of common viral diseases of cowpea (*Vigna unguiculata*) in a Southern Guinea savannah agro-ecology. *Asian Journal of Plant Pathology* 5:126–133. https://doi.org/10.3923/ajppaj.2011.126.133

Aruna, C., Rakshit, S., Shrotria, P.K., Pahuja, S.K., Jain, S.K., Siva Kumar, S., Modi, N.D., Deshmukh, N.T., Kapoor, R. and Patil, J.V. 2016. Assessing genotype by environment interactions and trait associations in forage sorghum using GGE biplot analysis. *The Journal of Agricultural Science* 154(1):73-86. https://doi.org/10.1017/S0021859615000106

Bashir, M., Ahmad, Z. and Ghafoor, A. 2002. Cowpea aphid-borne mosaic potyvirus: A review. *International Journal of Pest Management* 48(2):155-168. https://doi.org/10.1080/09670870110118722.

Blade, S.F., Shetty, S.V.R., Terao, T. and Singh, B.B. 1997. Recent developments in cowpea cropping systems research. In: Singh, B.B., Mohan Raj, D.R., Dashiel, K.E. and Jackai, L.E.N (Eds.), *Advances in cowpea research*. Co-publication of International Institute of Tropical Agriculture (IITA) and Japan International Research Center for Agricultural Sciences (JIRCAS), IITA, Ibadan, Nigeria. pp. 114-128.

Blanche, S.B., Myers, G. O. and Kang, M. S. 2007. GGE Biplots and Traditional Stability Measures for Interpreting Genotype by Environment Interactions. *Journal of Crop Improvement* 20(1):123-135. https://doi.org/10.1300/J411v20n01_07

Campbell, C.L. and Madden, L.V. 1990. Introduction to Plant Disease Epidemiology. John Wiley & Sons, New York, USA. 532pp.

Carsky, R.J., Vanlauwe, B. and Lyasse, O. 2002. Cowpea rotation as a resource management technology for cereal-based systems in the savannas of West Africa. In: Fatokun, C.A., Tarawali, S.A., Singh, B.B., Kormawa, P.M. and Tamò, M. (Eds.), *Challenges and opportunities for enhancing sustainable cowpea pro-duction*. Proceedings of the World Cowpea Conference III held at the International Institute of Tropical Agriculture (IITA), Ibadan, Nigeria, 4–8 September 2000. IITA, Ibadan, Nigeria. pp. 252-266.

Ceccarelli, S. 2012. Plant breeding with farmers –A technical manual. ICARDA, Aleppo, Syria. pp. xi + 126.

Edema, R., Adipala, E. and Florini, D. A. 1997. Influence of season and cropping system on occurrence of cowpea diseases in Uganda. *Plant Disease* 81:465-468.

Felipe de Mendiburu. 2017. agricolae: Statistical Procedures for Agricultural Research. R package version 1. 2-6.

https://CRAN.R-project.org/package=agricolae

Frutos, E., Galindo, M.P. and Leiva, V. 2014. An interactive biplot implementation in R for modeling genotype-by-environment interaction. *Stochastic Environmental Research and Risk Assessment* 28:1629-1641.

Goenaga, R., Quiles, A. and Gillaspie, A.G. 2008. Assessing yield potential of cowpea genotypes grown under virus pressure. *Hortscience* 43(3):673-676.

Gumedzoe, M.Y.D., Thottappilly, G. and Asselin, A. 1997. Occurrence of southern bean mosaic virus (SVMV) in Togo and its interaction with some cowpea cultivars. *African Crop Science Journal* 5(2):215-222.

Hawkes, J.G. 1991. The importance of genetic resources in plant breeding. *Biological Journal of the Linnean Society* 43(1):3-10.

Kone, N., Asare-Bediako, E., Silue, S., Kone, D., Koita, O., Menzel, W. and Winter, S. 2017a. Influence of planting date on incidence and severity of viral disease on cucurbits under field condition. *Annals of Agricultural Sciences* 62(1):99-104. https://doi.org/10.1016/j.aoas.2017.05.005.

Kone, N., Asare-Bediako, E., Koita, O., Kone, D. and Winter, S. 2017b. Seasonal and spatial variation in the prevalence of viral diseases and associated aphid-borne viruses in cucurbits in Cote d'Ivoire. *Annals of Agricultural Sciences* 62(2):227-234. https://doi.org/10.1016/j.aoas.2017.12.005

Mbeyagala, E.K., Mukasa, B.S., Tukamuhabwa, P. and Bisikwa, J. 2014. Evaluation of cowpea genotypes for virus resistance under natural conditions in Uganda. *Journal of Agricultural Science* 6(10):176–187. https://doi.org/10.5539/jas.v6n10p176

Ndiaye, M., Bashir, M., Keller, X.E. and Hampton, R.O. 1993. Cowpea viruses in Senegal, West Africa: Identities, distribution, seed transmission and sources of resistance. *Plant Disease* 77:999-1003.

Orawu, M., Melis, R., Milliano, W. D., Laing, M. and Adipala, E. 2005. Occurrence and prevalence of cowpea virus diseases in Uganda. *African Crop Science Conference Proceedings* 7:1279-1283.

Orawu, M., Obuo, J. and Omadi, R. 2015. Distribution and detection of cowpea viruses infecting cowpea in Uganda. *American Journal of Plant Sciences* 6(5):574-581. https://doi.org/10.4236/ajps.2015.65062

R Core Team, 2017. R. A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. URL: https://www.R-project.org/.

Taiwo, M.A., Hughes, J. d'A. and Oke, K.E. 2006. Studies on maize streak virus and Maize mottle/chlorotic stunt virus in Lagos, Nigeria. *Plant Disease* 90(2):199-202.

Taiwo, M.A. and Akinjogunla, O.J. 2006. Cowpea viruses: Quantitative and qualitative effects of single and mixed viral infections. *African Journal of Biotechnology* 5(19):1749-1756.

Timko, M. P., Ehlers, J. D. and Roberts, P. A. 2007. Cowpea (Chapter 3). In: Kole, C (Ed.), *Genome mapping and Molecular Breeding in plants* (Vol. 3). Berlin: Springer. pp. 50-67.

Timko, M.P. and Singh, B.B. 2008. Cowpea, a multifunctional legume (Chapter 10). In: In: Moore, P.H and Ming, R. (Eds.), *Genomics of Tropical Plants*. Berlin: Springer. pp. 227-258.

Yan, W. 2005. Use of biplot analysis in crop breeding. In: Swanson, C.S. (Ed.), *Proceedings of the Eastern Wheat Workers and Southern Small Grain Workers Conference*. May 9-12, 2005, Bowling Green, KY. University of Kentucky, USA. pp. 7-29.

Yan, W., Hunt, L. A., Sheng, Q. and Szlavnics, Z. 2000. Cultivar Evaluation and Mega-Environment Investigation based on the GGE Biplot. *Crop Science* 40:597-605.

Yan, W. and Rajcan, I. 2002. Biplot analysis of test sites and trait relations of soybean in Ontario. *Crop Science* 42:11-20.

Yan, W. and Tinker, N.A. 2005. An integrated Biplot analysis system for displaying, interpreting, and exploring genotype × environment interaction. *Crop Science* 45(3): 1004-1016. https://doi.org/10.2135/cropsci2004.0076

Yan, W. and Tinker, N. A. 2006. Biplot analysis of multi-environment trial data: Principles and applications. *Canadian Journal of Plant Science* 86(3):623-645.