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EFFECT OF *CUCUMBER MOSAIC VIRUS* INFECTED SEEDS ON GROWTH AND YIELD PARAMETERS OF SOYBEAN (*Glycinemax* L. MERR.)

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ABSTRACT

Cucumber mosaic virus (CMV) causes significant yield losses in legumes. To date, use of host-plant resistance remains the most effective and sustainable management approach. Experiment was conducted under screenhouse conditions to determine the effect of CMV-infected seeds on the growth and yield attributes of some selected soybean lines. Seeds of healthy soybean plants were planted as control. A total of twenty four soybean lines were evaluated using Completely Randomised Design with four replications. Disease incidence and severity, growth and yield attributes were recorded. Virus concentration in infected plants was determined at five weeks after sowing, using Antigen Coated Plate-Enzyme-Linked Immunosorbent Assay (ACP-ELISA). Data were subjected to Analysis of Variance (ANOVA). Growth and yield stability were determined using Additive Main Effects and Multiplicative Interaction (AMMI) analysis. All the seedlings from CMV-infected seeds elicited leaf chlorosis and mosaic symptoms of infection. At 5 weeks after sowing,TGX 1985-1D and TGX 1990-21F exhibited significantly (p<0.05) lowest disease severity score (symptom score = 2.3) whereas TGX 1995-5F had the highest severity score (symptom score = 4). The lowest virus titre (0.8) was found in TGX 1990-21Fwhile the highest (1.5) was observed in TGX 1989-63F. The effects ofsoybean lines and treatments (infected and control) accounted for 34.5 and 52.8 % of the total variation for 100-seed weight, respectively. The present study showed that TGX 1985-1D which exhibited significantly (p<0.05) lowest reductions in most of the parameters evaluated could be described as the most tolerant and utilized for breeding CMV-resistant soybean cultivars.

Keywords: *Cucumber mosaic virus*, Disease incidence, Disease severity, ELISA, Soybean, Yield

INTRODUCTION

Soybean is one of the major legume crops of global importance. It is very rich in essential amino acids for humans, and so is a good source of protein. In addition to its domestic uses, soybean is an essential ingredient in many processed foods, including dairy product substitutes (Kanchana

et al., 2016). Soybean, like most legumes, is capable of fixing nitrogen into the soil through a symbiotic relationship with the bacterium *Bradyrhizobium japonicum* (Bezdicek *et al*., 2008). Also, it is widely used in traditional medicine for prevention and control of human diseases (Afaf and Abdel-Had, 2014). The crop thrives well at mean temperatures of 23 to 25 ^{O}C on a wide range of soils, with optimum growth in moist alluvial soils having good organic matter content (Addo-Quaye *et al*., 2003). In 2013, about 6 million tonnes of soybeans were produced in Nigeria from 6 million hectares of land (FAO, 2013). Despite the large land area under cultivation yield per unit area is usually low (<1 t ha^{-1}) in tropical Africa. This has been attributed to biotic and abiotic factors. Abiotic factors include drought and high temperature (Oya *et al*., 2004; Thuzar *et al*., 2010). Economically important biotic constraints include diseases caused by *Pseudomonassyringae* pv. *glycinea*, and *Sclerotium rolfsii*(Rahayu, 2014).

Cucumber mosaic virus disease causes significant constraint to legume productivity (Hughes and Shoyinka, 2001). The disease is induced by *Cucumber mosaic virus* (CMV). *Cucumber mosaic virus* belongs to the genus *Cucumovirus* in the family *Bromoviridae* (Palukaitis and Garcia-Arenal, 2003). Symptoms of infection include mild to severe mottle, mosaic, distortion and reddish vein necrosis, depending on genetic background of the attacked plant (Agrios, 2005). Among the several control strategies use of tolerant varieties remains the most effective, ecologically sound, sustainable and cost-effective (Palukaitis and Garcia-Arenal, 2003). *Cucumber mosaic virus* is transmitted primarily by aphids, seed, and mechanically by humans. Over 60 species of aphid have been confirmed as insect vectors of the virus. These include *Aphis gossypii* Glover and *Myzus persicae* Sulzer (Alegbejo, 2015). Thevirus is transmitted in a non-persistent manner. Following acquisition by aphids in 5 – 10 seconds, it can be transmitted in less than one minute. Aphid's ability to transmit CMV declines after about 2 minutes and is usually lost within 2 hours. The virus induced mild mosaic and suppressed growth in susceptible soybean cultivars (Arogundade *et al.,* 2010).

Recently, there is a renewed interest to boost productivity of legumes including soybean through a package of appropriate inputs. However, any attempt to promote such activity must also take into consideration their level of resistance or at least tolerance to the prevailing virus diseases. Studies have shown that CMV is seed-borne and seed transmitted (O'keefe *et al*., 2007). In sub-Saharan Africa smallholder farmers usually rely on the seeds from previous harvest for field establishment in successive planting seasons. Seed transmission has been described as an effective means for plant virus survival and transmission over long distances. Therefore, the objective of this study was to determine the effect of CMV-infected seeds on the growth and yield attributes in some selected soybean lines.

MATERIALS AND METHODS

Study Location

The experiment was conducted under screenhouse conditions (temperature: 36 – 40 ^{o}C; relative humidity: 55 %) at the Teaching and Research Farm, Federal University of Technology (FUT), Minna (9^{0} 51'N, 6^{0} 44'E and 212 m above sea level), Niger State, Nigeria. Minna is located in the Southern Guinea Savanna with annual mean rainfall of 1200 mm. The rainfall is distributed between April and early October with peak around September. The relative humidity is between 40 and 60 % around January which later increases to between 60 and 80 % towards July.

Source of Planting Materials

Seeds harvested from CMV-infected soybean plants (Adamu *et al.*, 2015) were obtained from the stock in the Department of Crop Production, FUT. Minna. The seeds were harvested from diseased soybean plants and stored in paper bags for six months at room temperature in the laboratory. A total of twenty four soybean lines (TGX 1951-3F, TGX 1985-1D, TGX 1985-10F, TGX1987-10F, TGX 1987-62F, TGX 1988-5F, TGX 1989-19F, TGX 1989-62F, TGX 1989-63F, TGX 1989-69F, TGX 1989-1FN, TGX 1989-48FN, TGX 1989-49FN, TGX 1989-65FN, TGX 1989-68FN, TGX 1989-75FN, TGX 1990-3F, TGX 1990-21F, TGX 1990-67F, TGX 1991-10R, TGX 1993-5FN, TGX 1995-5F, TGX 2004-3F, TGX 2007-2F) were evaluated.

Treatments, Experimental Design and Crop Establishment

The experiment was laid out in Completely Randomized Design (CRD) with four replications. Soybean seeds were sown in plastic pots (30-cm diameter and 30-cm height) containing 1.5 kg heat sterilized (Agber and Adaikwu, 2012) loamy soil and seedlings were thinned to three plants per pot. Confirmed healthy soybean seeds were planted as control(Adamu *et al.*, 2015). All the plants were watered daily throughout the period of evaluation.

Observations and Data Collection

Plants were observed for disease incidence and severity. Disease incidence was taken as percentage of the seedlings showing symptoms of CMV infection at one and two weeks after sowing (WAS). Disease severity was recorded at 5 WAS, using a scoring scale of 1 – 5 (Arif and Hassan, 2002). In the scale: 1 = no symptoms (apparently healthy plant); 2 = slightly mosaic leaves (10 – 30 %); 3 = mosaic (31 – 50 %) and leaf distortion; 4 = severe mosaic (51 – 70 %), leaf distortion and stunting; 5 = severe mosaic (>70 %), stunting and death of plants. Number of leaves per plant, plant height and number of branches per plant were taken at 5 WAS. Number of flowers per plant, number of pods per plant, pod length, pod weight and 100-seed weight were also recorded.

Serological test

Leaves were collected from the plants at 5 WAS and analysed by Enzyme-Linked Immunosorbent Assay (ELISA) to confirm infection and virus concentration (Kumar, 2009). Eachsample was homogenized in coating buffer at the rate of 100 mg/mL. This was followed by addition of 100 µL of each sample into duplicate wells of the ELISA plate. The plate was incubated at 37 ^{O}C for 1 hour, washed thrice with Phosphate Buffered Saline-Tween (PBS-T) and tap-dried on a paper towel. One gram of healthy soybean leaf was ground in 20 mL of conjugate buffer. Rabbit antibody for CMV diluted with conjugate buffer (1:10, 000) was added at the rate of 100 µL per sample. The plate was incubated at 37 ^{O}C for 1 hour, washed thrice with PBS-T and tap-dried. This was followed by addition of 100 µL of anti-rabbit goat antimouse diluted with conjugate buffer at the rate of 1:15,000 dilutions. Moreover, the plate was incubated at 37 ^{O}C for 1 hour and washed with PBS-T. Substrate solution was added at the rate of 100 µL per sample. Absorbance readings were recorded at 405 nm using a microplate reader (MRX, Dynex Technologies, Inc., USA) after overnight incubation of the plate in the dark at room temperature. Values were considered to be positive when the mean absorbance reading was at least twice that of the mean for the negative control.

Statistical Analysis

Data were subjected to Analysis of Variance (ANOVA) and significance was determined at $p \leq 0.05$. Where ANOVA results were significant the difference between infected and healthy plants was compared using the Least Significant Difference (LSD). Growth and yield stability under CMV infection and disease-free conditions were verified using Additive Main effects and Multiplicative Interactions (AMMI) analysis according to McDermott and Coe (2012). Statistical analysis was performed using Breeding Management System (BMS, 2015).

RESULTS

Disease Incidence and Severity

All the seedlings from CMV-infected seeds elicited symptoms of CMV disease, whereas seedlings from healthy seeds were apparently symptomless (symptom score = 1). Symptoms first appeared at 2 weeks after seedling emergence. Symptoms started as mild mottling of the topmost leaves. At 3 weeks after emergence, some plants exhibited mosaic and leaf curling symptoms. Disease severity differed signifIcantly ($p<0.05$) among the soybean lines. At 5 WAS, disease severity varied significantly ($p<0.05$) between 2.3 and 4 (Fig. 1A) with the lowest observed in TGX 1985-1D(symptom score = 2.3) and TGX 1990-21F (symptom score = 2.3), while the highest value was found in TGX 1995-5F (symptom score = 4). Moreover, 66.7 % (TGX 1951-3F, TGX1987-10F, TGX 1987-62F, TGX 1988-5F, TGX 1989-19F, TGX1989-62F, TGX 1989-69F, TGX 1989-1FN, TGX 1989-65FN, TGX 1989-68FN, TGX 1990-3F, TGX 1990-67F, TGX 1991-10R, TGX 1993-5FN,. TGX 2004-3F and TGX 2007-2F) of the soybean lines had a severity score of 3. Conversely, in TGX 1989-63F, TGX 1989-48FN and TGX 1989-75FN

severity scores averaged 3.3. Furthermore, symptom score of 3.7 was observed in TGX 1989-49FN. All the leaves from virus-infected plants tested positive for CMV in ELISA whereas the healthy (control) plants showed negative reactions (Fig. 1B). The differences in virus concentrations among the infected plants were significant ($p<0.05$) and the trend was as reported for symptom severity scores.

Growth and Yield Parameters

Number of leaves per plant varied between 28 (TGX 1988-5F) and 49 (TGX 2004-3F) in the healthy plants. On the other hand, number of leaves per plant ranged from 8 (TGX 1990-3F) to 22 (TGX 1987-62F) in the CMV-infected plants. Tables 1 and 2 show the growth and yield data obtained from healthy and CMV-infected soybean plants. Healthy plants were significantly ($p<0.05$) taller than the CMV-infected plants.Healthy plants showed rapid and normal growth while infected plants exhibited slow growth with reduced heights. The heights of healthy plants varied from 58.3 (TGX 1987-62F) to 76.8 cm (TGX 1989-68FN), which differed from 39.3 (TGX 1989-62F) to 53.4 cm (TGX 2004-3F) observed in the CMV-infected plants.

Healthy plants produced branches ranging from 12 (TGX 1990-67F, TGX 1990-3F and TGX 1993-5FN) to 18 (TGX 1985-10F, TGX 1989-1FN, TGX 1989-49FN and TGX 1989-65FN) per plant whereas number of branches per plant varied between 6 (TGX 1995-5FN) and 12 (TGX 1985-10F) in the CMV-infected plants. Again, the difference between the two categories was significant ($p<0.05$) in all the soybean lines. Number of flowers per plant in healthy plants varied between 6 (TGX 1989-1FN) and 31 (TGX 1987-62F) compared to the CMV-infected plants which had a range of 4 (TGX 1989-1FN, TGX 1989-48FN and TGX 1989-49FN) to 19 (TGX 1987-62F) flowers per plant. *Cucumber mosaic virus* disease significantly ($p<0.05$) affected the reproductive performance of infected plants. Healthy plants produced more pods per plant with values ranging between 18 (TGX 1990-3F) and 37 (TGX 1989-65FN) per plant than their CMV-infected counterparts which had 8 (TGX 1995-5FN) to 24 (TGX 1985-10F) pods per plant, and the differences were statistically significant ($p<0.05$) in all the soybean lines. Similarly, the pods from healthy plants were generally longer with values varying from 4 (TGX 1989-62F) to 5.7 cm (TGX 2007-2F) contrary to the CMV-infected plants which had a range of 2.7 (TGX 1995-5FN) to 4.3 cm (TGX 1989-65FN), and the differences were significant ($p<0.05$). In the same vein, healthy plants produced heavier pods which ranged between 1.2 (TGX 2004-3F) and 2.1 g (TGX 1985-10F) per plant compared to their CMV-infected plants which exhibited 0.9 (TGX 1988-5F) to 1.5 g (TGX 1985-10F). One hundred-seed weights of the healthy plants varied between 9.2 (TGX 1989-62F) and 13.7 g (TGX 1995-5FN) while in the CMV-infected plants values ranged from 7.5 (TGX 1987-62F) to 10.1 g (TGX 1985-10F). Narrow sense heritability for growth and yield characters varied between 1 and 68 % among the CMV-infected plants, which was different from a range of 19 to 81 % observed in the healthy plants.

Lapai Journal of Science and Technology, Vol. 4, No. 1 (2018)

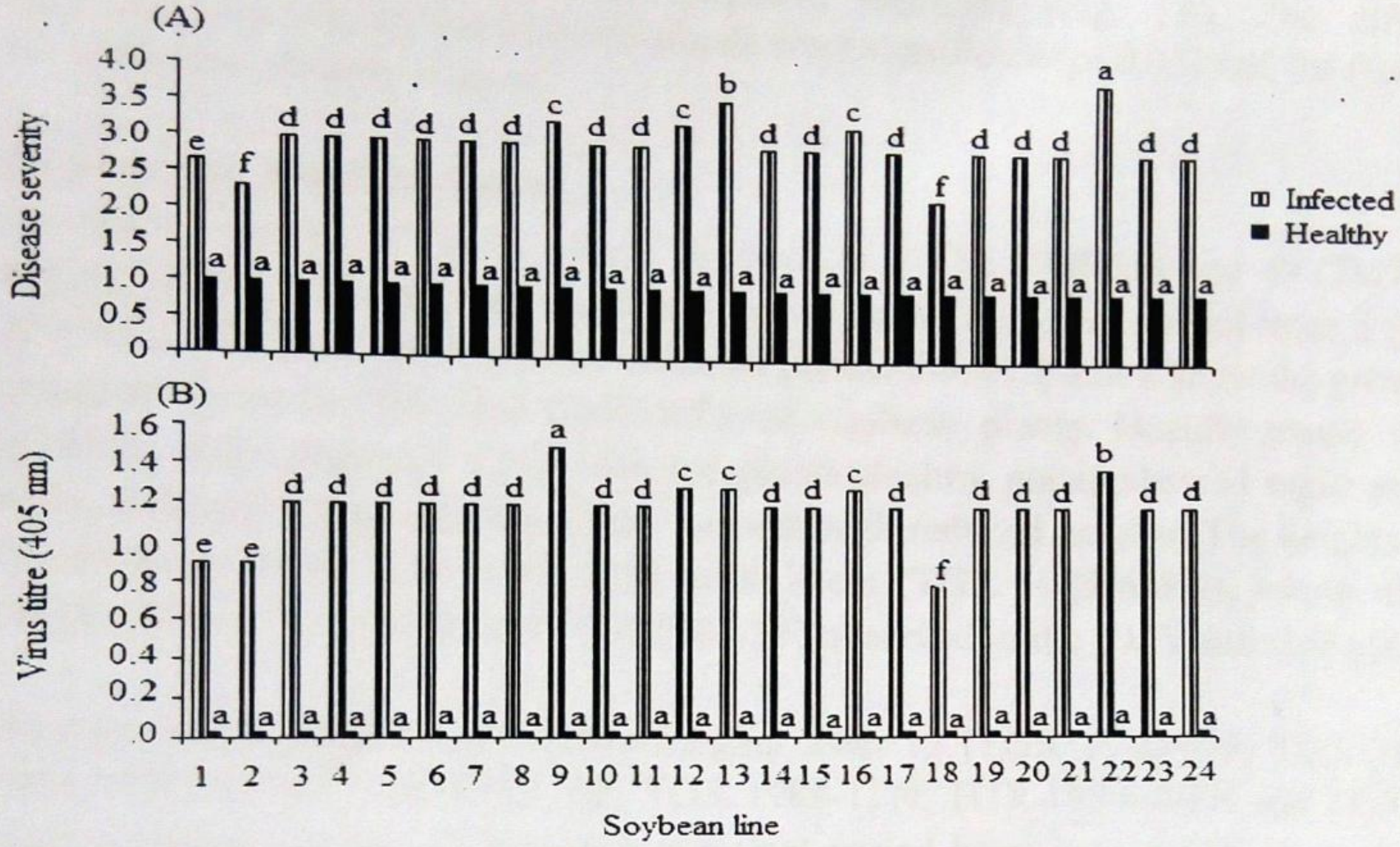

Fig. 1: Symptom scores (A) and virus concentrations (B) of the CMV –infected and healthy soybean plants at 5 weeks after sowing in a screenhouse; The means of each treatment labelled with dissimilar letter differ significantly ($p \leq 0.05$) according to Duncan Multiple Range Test (DMRT)

Key: 1 = TGX 1951-3F, 2 = TGX 1985-1D, 3 = TGX 1985-10F, 4 = TGX1987-10F, 5 = TGX 1987- 62F, 6 = TGX 1988-5F, 7 = TGX 1989-19F, 8 = TGX 1989-62F, 9 = TGX 1989-63F, 10 = TGX 1989-69F, 11 = TGX 1989-1FN, 12 = TGX 1989-48FN, 13 = TGX 1989-49FN, 14 = TGX 1989- 65FN, 15 = TGX 1989-68FN, 16 = TGX 1989-75FN, 17 = TGX 1990-3F, 18 =TGX 1990-21F, 19 = TGX 1990-67F, 20 = TGX 1991-10R, 21 = TGX 1993-5FN, 22 = TGX 1995-5F, 23 = TGX 2004-3F, 24 = TGX 2007-2F

Table 1: Effect of CMV infection on growth parameters of soybean plants

Soybean Line	Leaves per plant (no.)		Plant height (cm)		Branches per plant (no.)		Flowers per plant (no.)	
	Infected	Healthy	Infected	Healthy	Infected	Healthy	Infected	Healthy
TGX 1951-3F	11[b]	38[a]	48.9[b]	67.5[a]	10[b]	15[a]	7[b]	10[a]
TGX 1985-1D	18[b]	34[a]	46.5[b]	68.0[a]	11[b]	16[a]	10[b]	13[a]
TGX 1985-10F	17[b]	44[a]	49.6[b]	66.4[a]	12[b]	18[a]	15[b]	19[a]
TGX 1987-10F	12[b]	31[a]	47.6[b]	65.9[a]	8[b]	13[a]	7[b]	10[a]
TGX 1987-62F	22[b]	45[a]	47.6[b]	58.3[a]	8[b]	17[a]	19[b]	31[a]
TGX 1988-5F	12[b]	28[a]	47.6[b]	70.9[a]	7[b]	14[a]	5[b]	9[a]
TGX 1989-19F	13[b]	42[a]	46.2[b]	64.0[a]	7[b]	14[a]	12[b]	17[a]
TGX 1989-62F	14[b]	43[a]	39.3[b]	66.2[a]	9[b]	14[a]	15[b]	23[a]
TGX 1989-63F	15[b]	36[a]	44.5[b]	62.9[a]	9[b]	14[a]	8[b]	18[a]
TGX 1989-69F	16[b]	46[a]	45.8[b]	68.0[a]	8[b]	16[a]	12[b]	24[a]
TGX 1989-1FN	17[b]	44[a]	49.1[b]	71.0[a]	8[b]	18[a]	4[b]	6[a]
TGX 1989-48FN	20[b]	44[a]	45.5[b]	70.0[a]	9[b]	17[a]	4[b]	8[a]
TGX 1989-49FN	12[b]	40[a]	46.2[b]	65.4[a]	8[b]	18[a]	4[b]	10[a]
TGX 1989-65FN	15[b]	40[a]	45.6[b]	73.2[a]	9[b]	18[a]	12[b]	18[a]
TGX 1989-68FN	12[b]	33[a]	52.1[b]	76.8[a]	9[b]	16[a]	11[b]	16[a]
TGX 1989-75FN	11[b]	34[a]	41.8[b]	62.4[a]	7[b]	14[a]	9[b]	14[a]
TGX 1990-3F	8[b]	32[a]	40.5[b]	62.4[a]	7[b]	12[a]	6[b]	8[a]
TGX 1990-21F	16[b]	40[a]	46.3[b]	66.1[a]	8[b]	14[a]	5[b]	7[a]
TGX 1990-67F	17[b]	36[a]	48.3[b]	64.3[a]	8[b]	12[a]	6[b]	8[a]
TGX 1991-10R	16[b]	43[a]	45.7[b]	63.7[a]	8[b]	15[a]	14[b]	20[a]
TGX 1993-5FN	15[b]	42[a]	48.5[b]	73.8[a]	7[b]	12[a]	7[b]	8[a]
TGX 1995-5FN	15[b]	41[a]	42.1[b]	69.3[a]	6[b]	16[a]	6[b]	13[a]
TGX 2004-3F	18[b]	49[a]	53.4[b]	73.0[a]	7[b]	16[a]	7[b]	16[a]
TGX 2007-2F	13[b]	45[a]	41.0[b]	64.2[a]	10[b]	17[a]	12[b]	18[a]
Heritability	0.05	0.34	0.01	0.51	0.30	0.71	0,68	0.81
p-value	0.11	0.42	0.48	0.02	0.00	0.14	0.00	0.00

Means of each soybean line with dissimilar letters within the row differ significantly ($p \leq 0.05$) by Least Significant Difference (LSD)

Table 2: Effect of CMV infection on yield parameters of soybean plants

Soybean Line	Pod per plant (no.)		Pod length (cm)		Pod weight (g)		100-seed weight (g)	
	Infected	Healthy	Infected	Healthy	Infected	Healthy	Infected	Health
TGX 1951-3F	22[b]	30[a]	4.0[b]	5.3[a]	1.2	1.6	9.9	11.7
TGX 1985-1D	19[b]	27[a]	3.3[b]	5.3[a]	1.4	1.6	9.5	10.1
TGX 1985-10F	24[b]	28[a]	3.0[b]	5.0[a]	1.5	2.1	10.1	11.2
TGX 1987-10F	13[b]	27[a]	3.7[b]	5.3[a]	1.3	1.6	8.4	11.2
TGX 1987-62F	21[b]	34[a]	3.3[b]	5.0[a]	1.1	1.7	7.5	9.7
TGX 1988-5F	13[b]	21[a]	3.3[b]	5.0[a]	0.9	1.4	8.8	10.2
TGX 1989-19F	13[b]	25[a]	3.3[b]	5.7[a]	1.3	1.6	9.1	10.1
TGX 1989-62F	23[b]	34[a]	3.3[b]	4.0[a]	1.2	1.6	8.3	9.2
TGX 1989-63F	8[b]	28[a]	3.3[b]	5.3[a]	1.2	1.6	9.2	10.5
TGX 1989-69F	19[b]	33[a]	3.0[b]	5.3[a]	1.5	1.8	8.6	9.6
TGX 1989-1FN	19[b]	27[a]	3.0[b]	5.3[a]	1.4	1.7	9.2	11.8
TGX 1989-48FN	18[b]	31[a]	3.3[b]	5.3[a]	1.0	1.3	9.3	10.9
TGX 1989-49FN	12[b]	28[a]	4.0[b]	5.3[a]	1.2	1.4	9.7	10.8
TGX 1989-65FN	20[b]	37[a]	4.3[b]	5.3[a]	1.4	1.9	8.7	9.9
TGX 1989-68FN	18[b]	29[a]	3.7[b]	5.3[a]	1.1	1.4	8.7	10.7
TGX 1989-75FN	15[b]	25[a]	3.7[b]	5.3[a]	1.1	1.3	9.3	10.4
TGX 1990-3F	12[b]	18[a]	3.3[b]	5.7[a]	1.2	1.5	9.6	11.8
TGX 1990-21F	13[b]	23[a]	4.0[b]	4.3[a]	1.3	1.9	9.3	12.3
TGX 1990-67F	15[b]	28[a]	3.7[b]	5.0[a]	1.2	1.8	9.8	11.7
TGX 1991-10R	20[b]	31[a]	3.0[b]	4.7[a]	1.2	1.5	9.4	10.7
TGX 1993-5FN	21[b]	29[a]	4.0[b]	5.0[a]	1.2	1.8	9.6	11.5
TGX 1995-5FN	8[b]	28[a]	2.7[b]	5.0[a]	1.1	1.8	9.2	13.7
TGX 2004-3F	15[b]	28[a]	3.7[b]	5.0[a]	1.1	1.2	9.9	10.8
TGX 2007-2F	21[b]	33[a]	3.3[b]	5.7[a]	1.3	1.8	9.1	10.5
Heritability	0.50	0.54	0.19	0.38	0.39	0.58	0.11	0.42
p-value	0.01	0.02	0.08	0.27	0.01	0.07	0.36	0.06

Means ofeach soybean line with dissimilar letters within the row differ significantly ($p \leq 0.05$) by Least Significant Difference (LSD)

Growth and Yield Stability

The effect of soybean lines was highly significant (p<0.01) for all the parameters evaluated except for number of leaves per plant and pod length (Table 3). Conversely; the effect of treatments was highly significant (p<0.01) for all the parameters evaluated. Additive Main effects and Multiplicative Interaction analysis (Table 4) revealed that variation due to effect of treatments was consistently higher than effect of the soybean lines except for number of flowers per plant. In number of leaves per plant, the effect of soybean lines and treatments accounted for 14.9 and 81.1 % variation, respectively. In plant height, the effect of soybean lines and treatments induced 1 and 97.9 % variation, respectively. Variation in number of branches per plant due to the effect of soybean lines and treatments was 12 and 82.7 %, respectively. Furthermore, the effect of soybean lines resulted in higher (74 %) variability than treatments (19.5 %) effect with respect to number of flowers per plant.

The effect of soybean lines accounted for 28 % variation in number of pods per plant which was lower than 64.8 % induced by the treatments. While only 9 % of the total variation was due to effect of soybean lines on pod length, treatments' effect accounted for 82.3 % of the total variation.. In pod weight per plant, the effect of soybean lines caused 38.5 % of the observed variation while 54.9 % was due to the treatments. Variation resulting from effect of soybean lines and treatments for 100-seed weight was 34.5 and 52.8 % of the total, respectively. None of the soybean lines exhibited consistent stability. However, TGX 1985-1D was the most stable soybean line for number of leaves per plant and 100-seed weight (Table 5). Other soybean lines such as TGX 1987-62F, TGX 1990-67F, TGX 1989-1FN, TGX 1985-10F, TGX 1990-21F, and TGX 2004-3F expressed the highest level of stability for plant height, number of branches per plant, number of flowers per plant, number of pods per plant, pod length, and pod weight per plant, respectively.

Table 3: Mean squares from AMMI analysis of the growth and yield characters in CMV-infected and healthy soybean plants

Source of variation	DF	Leaves per plant (no.) Mean squares	Plant height (cm) .Mean squares
Soybean lines	23	114.9	69.6**
Treatments	1	14346.7**	15880.2**
Interaction	23	30.1	23.9
Residuals	96	77.3	33.7

Source of variation	DF	Branches per plant (no.)	Flowers per plant (no.)

Soybean lines	23	12.3**	157**
Treatments	1	1736.1**	949**
Interaction	23	4.8	13.8
Residuals	96	4.9	23.4
		Pods per plant (no.)	Pod length (cm)
Soybean lines	23	92.8**	0.4
Treatments	1	4935.1**	97.7**
Interaction	23	23.7	0.4
Residuals	96	28	0.3
		Pod weight (g)	100-seed weight (g)
Soybean lines	23	0.2**	2.9**
Treatments	1	5.7**	103.7**
Interaction	23	0.0	1.1
Residuals	96	0.1	1.4

**Significant at p=0.01

Table 4: Additive Main Effects and Multiplicative Interaction (AMMI) analysis of variance for growth and yield characters in CMV - infected and healthy soybean plants

Source of variation	DF	Leaves per plant (no.)		Plant height (cm)		Branches per plant (no.)		Flowers per plant (no.)	
		Sum of squares	Mean squares	Sum of squares	Mean squares	Sum of squares	Mean squares	Sum of squares	Mean Squares
Soybean lines	23	7926.0	345.0	56.7	2.5	94.1	4.1	10833.0	471.0
Treatments	1	43040.0	43040.0	5293.4	5293.4	578.7	578.7	2847.0	2847.0
Interactions	23	2078.0	90.0	56.7	2.5	36.7	1.6	952.0	41.4
IPCA 1	23	2078.0	90.0	56.7	2.5	36.7	1.6	952.0	41.4
IPCA 2	21	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Residuals	–21	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
		Pods per plant (no.)		Pod length (cm)		Pod weight per plant (g)		100-seed weight per plant (g)	
Soybean lines	23	711.7	30.9	3.7	0.2	1.3	0.1	23.0	1.0
Treatments	1	1645.0	1645.0	33.9	33.9	1.9	1.9	34.6	34.6
Interactions	23	181.9	7.9	3.6	0.2	0.2	0.0	8.3	0.4
IPCA 1	23	181.9	7.9	3.6	0.2	0.2	0.0	8.3	0.4
IPCA 2	21	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Residuals	–21	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0

Table 5: Top 10 soybean sensitivity (stability) coefficients for growth and yield characters of CMV –infected soybean plants

Leaves per plant (no.)		Plant height (cm)		Branches per plant (no.)		Flowers per plant (no.)	
Soybean ID	Sensitivity	Soybean ID	Sensitivity	Soybean ID	Sensitivity	Soybean ID	Sensitivity
2	0.5399	5	0.7846	19	0.5760	11	0.2597
6	0.6067	16	0.8822	9	0.6720	19	0.3030
19	0.6846	17	0.8822	17	0.7200	21	0.3679
5	0.7403	9	0.8951	21	0.7680	18	0.4112
4	0.7458	20	0.9136	8	0.7680	1	0.4977
15	0.8349	7	0.9209	1	0.7680	17	0.5194
9	0.8516	24	0.9274	18	0.7680	2	0.5627
12	0.8627	19	0.9282	3	0.7680	12	0.6492
18	0.9573	13	0.9564	2	0.8160	4	0.6492
16	0.9573	4	0.9669	4	0.8160	3	0.6709
Pod per plant (no.)		Pod length (cm)		Pod weight per plant (g)		100-seed weight per plant (g)	
3	0.3986	18	0.1983	23	0.2921	2	0.3535
17	0.4840	8	0.3967	16	0.4673	23	0.5342
11	0.6548	21	0.5950	2	0.6342	8	0.5578
2	0.6548	14	0.5950	13	0.6426	7	0.5912
6	0.7117	23	0.7934	12	0.6509	10	0.6147
21	0.7117	13	0.7934	7	0.7594	13	0.6187
1	0.7402	19	0.7934	4	0.7928	16	0.6619
18	0.8826	1	0.7934	17	0.8178	3	0.6717
8	0.8826	4	0.9917	1	0.8428	14	0.7385
16	0.9110	16	0.9917	15	0.8762	20	0.7542

Key: 1 = TGX 1951-3F, 2 = TGX 1985-1D, 3 = TGX 1985-10F, 4 = TGX1987-10F, 5 = TGX 1987-62F, 6 = TGX 1988-5F, 7 = TGX 1989-19F, 8 = TGX 1989-62F, 9 = TGX 1989-63F, 10 = TGX 1989-69F, 11 = TGX 1989-1FN, 12 = TGX 1989-48FN, 13 = TGX 1989-49FN, 14 = TGX 1989-65FN, 15 = TGX 1989-68FN, 16 = TGX 1989-75FN, 17 = TGX 1990-3F, 18 = TGX 1990-21F, 19 = TGX 1990-67F, 20 = TGX 1991-10R, 21 = TGX 1993-5FN, 22 = TGX 1995-5F, 23 = TGX 2004-3F, 24 = TGX 2007-2F

DISCUSSION

The development of typical symptoms of CMV on the seedlings of CMV-infected seeds indicated that the virus could survive in planting material between seasons. This result supported the earlier report that the virus was seed-borne (Alegbejo, 2015) and also transmitted by infected seeds.

Survival in seeds of susceptible plants has been described as one of the several avenues facilitating the spread of plant viruses between seasons (Sevik and Kose-Tohumcu, 2011). The symptoms observed on the leaves of CMV-infected plants were in agreement with those earlier reported (Hughes and Shoyinka, 2001). TGX 1985-10F and TGX 1990-21F which exhibited the lowest disease severity score at 5 WAS could be described as the most tolerant lines whereas TGX 1995-5F which had the highest disease severity score was the most susceptible to CMV infection. Furthermore, since the symptom scores followed a trend similar to ELISA results, there was a positive correlation between visual disease assessment and serological test. These results confirmed the findings of Maruthi *et al.* (2003) who demonstrated a positive relationship between visual severity scores and DNA concentration in tomato plants infected with *Tomato yellow leaf curl virus*.

Higher growth and yield values were obtained in the healthy plants than those infected with CMV owing to the deleterious impact of the pathogen. This corroborated the results documented by Arogundade *et al.* (2010) who encountered considerable reductions in soybean plants infected with CMV. Viruses normally hijack the physiological activities of susceptible plants for self replication and establishment. Therefore, the activity of important organ such as chloroplast was probably inhibited in infected plants. Studies have shown that retardation of chloroplast could suppressphotosynthesis in diseased plant (Agrios, 2005). The deleterious effect of infection was also probably due to inability of the roots of diseased plants to absorb sufficient nutrients and moisture from the soil. Impairment of plant physiology possibly culminated in poor growth and reduced yield (Agrios, 2005). Expression of substantially higher heritability in healthy plants than the CMV-infected was probably because virus infection masked full expression of the growth and yield potentials in the diseased plants.

The differences in response of the soybean lines to CMV disease were could be attributable to the variability in their genetic backgrounds. This agreed with the observations documented when some maize cultivars were infected with *Maize streak virus* (MSV) (Salaudeen *et al.*, 2011). The fact that healthy plants consistently exhibited higher growth and yield attributes than infected plants in most of the growth and yield parameters supported the findings of Adama*et al.* (2015) when some soybean lines were investigated under *Blackeye* cowpea *mosaicvirus* (BlCMV) infection and diseases free conditions. This indicated that CMV-infected soybean seeds are not suitable for plant regeneration. The soybean line TGX 1985-1D which expressed significantly ($p<0.05$) highest level of stability revealed that its growth and yield parameters were mildly impaired under CMV disease condition. This implied that its general performance could be predicted and would be useful in breeding for resistance against CMV disease. However, TGX 1987-62F, TGX 1990-67F, TGX 1989-1FN, TGX 1985-10F, TGX 1990-21F, and TGX 2004-3F which exhibited the highest level of stability for plant height, number of branches per plant, number of flowers per plant, number of pods per plant, pod length, and pod weight per plant could be utilized for various breeding purposes. The AMMI analysis indicating consistently higher effect of treatments than soybean lines implied that CMV-free soybean seeds should be planted in order to guarantee appreciable yield.

CONCLUSION

This investigation revealed the deleterious effect of CMV on susceptible soybean lines. TGX 1985-1D and TGX 1990-21F exhibited the lowest disease severity scores whereas TGX 1995-5F was the most vulnerable. However, TGX 1985-1D which exhibited significantly ($p<0.05$) lowest reductions in most of the parameters evaluated could be described as the most tolerant and utilized for breeding CMV-resistant soybean cultivars.

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