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INCIDENCE AND SEVERITY OF FUNGAL AND COMMON VIRAL DISEASES OF SOME SOYBEAN LINES IN A DERIVED GUINEA SAVANNAH AGRO-ECOLOGY

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Abstract: Nigeria is the largest producer and consumer of soybean in Sub-Saharan Africa with a low yield of less than 1 ton per hectare. Plant diseases play a major role in yield reduction for the crop. The study was to investigate the incidence and severity of fungal and common viral diseases of some soybean lines and determine their effects on soybean yield. Nineteen newly developed soybean lines with two local checks were evaluated. Fungal isolates were identified using cultural and morphological characteristics while Antigen Coated Plate-Enzyme-Linked Immunosorbent Assay was used for detecting viruses. Data were subjected to Analysis of Variance and means were separated at $P \leq 0.05$ using Duncan's Multiple Range Test. Eight fungi isolated from diseased soybean plants were *Fusarium oxysporum*, *Choanephora infundibulifera*, *Colletotrichum gloeosporioides*, *Culvularia* spp., *Fusarium verticilloides*, *Aspergillus flavus*, *Lasiodiplodia theobromae* and *Pestalotia* spp., while the common viral symptom on the field was mosaic and mottling. *F. oxysporum* recorded the highest frequency of occurrence of 40.91% and 22.30%, in both years. *F. oxysporum* and *C. infundibulifera* showed characteristics symptoms of blight when used for pathogenicity on both checks. The soybean lines differed significantly [$P \leq 0.05$] in disease incidence and severity for both fungi and viral diseases. All lines were positive for Cowpea mild mottle virus [CPMMV] in 2016. All the lines evaluated were moderately resistant to leaf blight disease. The study concluded that these lines were tolerant to all observable diseases occasioned by their abilities to produce higher grain yield compared with the local checks, despite the high disease incidence and severity.

Keywords: Cowpea mild mottle virus, *Fusarium* blight, *Fusarium oxysporum*, incidence and severity, leaf blight, resistant.

Introduction

Soybean (*Glycine max* L.) is one of the most important oilseeds crop all over the world [ANONYMOUS, 2018] widely cultivated in tropical, subtropical, and temperate climates of the world [IITA, 2009]. The spread of the crop from its native land of origin has been mainly due to its adaptability and predominant use as a food crop for human nutrition, source of protein for animals, medicinal plant and lately as an industrial crop [YUSUF & IDOWU, 2001]. This legume provides cheap and high-quality protein, containing all amino acids essential for human nutrition when compare to meat and eggs. The crop can be successfully grown in many states of the country, using low agricultural input. Its cultivation

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in Nigeria has expanded as a result of its nutritive and economic importance and diverse domestic usage. Soybean has an average protein content of 40% and is more protein-rich than any of the common vegetable or animal food sources found in Nigeria. When oil is extracted from soybean the residue left is used as protein supplement in livestock feeds. Soybean seeds also contain about 20% oil on a dry matter basis, and this is 85% unsaturated and cholesterol-free [DUGJE & al. 2009].

Nigeria is the largest producer and consumer of soybean in Sub-Saharan Africa with a low yield of less than 1 ton per hectare [IITA, 2009]. Several factors are attributed to this low yield, such as climatic conditions, differences in rainfall patterns, outbreak of diseases and pests etc. Among these factors, plant diseases play a major role in yield reduction for the crop. The increase in the number of soybean diseases and their expansion emanate from intensive production and increased acreage in new regions of the world [HARTMAN & al. 2005]. More than 300 diseases have been reported to affect soybean worldwide [HARTMAN & al. 1999; HARTMAN & al. 2005]. All parts of soybean plant are susceptible to a number of pathogens which reduce quality and/or quantity of seed yield, due to the facts that propagules of various pathogens have increased to densities that cause economic yield losses [HARTMAN & al. 2005].

Among the devastated diseases of soybean are, *Fusarium* blight or wilt disease of soybean, caused by the common soil-borne fungus *Fusarium oxysporum*; it is one of the most destructive diseases of soybean [HASHEM & al. 2009; FAYZALLA & al. 2009]. The pathogen can affect soybeans at any stage of development [FERRANT & CARROLL, 1981]. *F. oxysporum* can also cause root rot and wilt disease of soybean [RAHMAN & al. 2020] *Fusarium* blight symptoms are more noticeable under reduced moisture and hot conditions. The pathogen is difficult to control owing to its persistence nature in the soil and wide host range [ABDEL-MONAIM & al. 2011]. *Phytophthora sojae* causes seedling blight, root and stem rot, this disease is rapidly becoming a very destructive disease in Nigeria [DUGJE & al. 2009]. Asian soybean rust, caused by *Phakopsora pachyrhizi*, is another important soybean foliar disease in Nigeria. The infected leaves have small tan to dark brown or reddish-brown lesions on which small raised pustules occur on the lower surface of the leaves, severe infection leads to premature defoliation and yield losses up to 80% had been recorded [DUGJE & al. 2009]. The crop is also, susceptible to several viruses transmitted by aphids, beetles and whiteflies prevailing in Nigeria, Cowpea mild mottle virus (CPMMV; genus *Carlavirus* transmitted by whitefly (*Bemisia tabaci*) is the most prevalent virus associated with soybean mosaic disease in Nigeria [DUGJE & al. 2009]. Infection with Soybean mosaic virus (SMV) causes mosaic symptoms (light and dark green areas, chlorosis, and leaf curl), necrosis (necrotic areas, stem browning, and stem tip necrosis), and seed mottling, resulting in serious yield losses [ZHENG & al. 2005], yield losses due to SMV infection range from 8% to 50% under natural field conditions [HILL, 1999], to total crop loss during severe outbreaks [LIAO & al. 2002].

Hence, the objectives of this study were, to investigate the incidence and severity of fungal and common viral diseases of some soybean lines, confirm the pathogenicity of the isolates and to determine their effects on soybean yield.

Materials and methods

Experimental site

This study was carried out during the 2015 late cropping season and 2016 early cropping season at Research Farm of National Cereal Research Institutes, Ibadan Research Station, Latitude 7º22' N and Longitude 3º58' E with mean annual rainfall of 1150-1250 mm.

Soybean Lines used

The lines are Early lines: TGx 1990-40F, TGx 1989-48FN, TGx 1989-68FN, TGx 1990-55F, TGx 1989-40F, TGx 1990-52F, TGx 1989-49FN, TGx 1990-57F, TGx 1990-55F, TGx 1485-1D (Check). Medium lines: TGx 1989-45F, TGx 1989-11F, TGx 1989-75FN, TGx 1990-114FN, TGx 1990-78F, TGx 1993-4FN, TGx 1989-53FN, TGx 1990-95F, TGx 1989-42F, TGx 1990-110FN, TGx 1448-2E (Check), and were collected from International Institute for Tropical Agriculture (IITA), Ibadan.

Experimental design and disease assessment

The lines were laid out in a Randomized Complete Block Design with three replications. Plot sizes were 4m row length with inter-row spacing of 50cm and 5cm intra-row, and were observed for natural development of foliar diseases symptoms. Soybean leaf blight severities were determined according to [ABDOU & al. 2001] using rating scale of 1-5:1 = no yellow/spots on leaf, 2 = (1-25%) yellow colouration on one leaf, 3 = (26-50%) yellow colouration on more than one leaf, 4 = (51-75%) yellow colouration plus one wilted leaf, 5 = (76-100%) yellow colouration with more than one wilted leaf. While virus disease severities on the different plots were assessed using a modified scale of 1-5, by ASADI (2005): 1 = no visible symptoms, 2 = mild leaf mottling, 3 = chlorosis and mottling, 4 = stunted with severe mottling and chlorosis, 5 = stunted, severe mottling, leaf bunching, chlorosis with leaf defoliation. Disease incidence was determined by counting diseased plants and expressing it as a percentage of total plants in each plot.

Resistance level

The plants were rated as tolerant, resistant or susceptible on the basis of the following scales: resistance or otherwise to fungal diseases were assessed according to [EL-BRAMAWY & ABD AL-WAHID, 2009], using a scale of 1-5 based on the % disease incidence of: 0.1-20% resistant (R), 20.1-40% moderately resistant (MR), 40.1-50% moderately susceptible (MS), 50.1-75% susceptible (S), 75.1-100% highly susceptible (HS). And soybean viral resistances were assessed based on the mean severity, using modified scale of 1-5 by AKBAR & al. (2015): 1 = (1.0-1.9) highly resistant (H), 2 = (2.0-2.99) moderately resistant (MR), 3 = (3.0-3.99) moderately susceptible (MS), 4 = (4.0-4.99) susceptible (S), 5 = (5 and above) highly susceptible (HS).

Isolation and identification of pathogen associated with soybean foliar diseases

Potato dextrose agar was used for fungal cultures, by dissolving thirty-nine grams of the agar in 1000mls of distilled water and autoclaved at 121°C for 15 minutes, allowed to cool sufficiently before pouring on Petri dishes, 2 mm of the diseased sample, surface sterilized in 3% sodium hypochlorite solution for 1 minute, rinsed in sterile distilled water and then dried in three folds of Whatman's filter paper was then inoculated on the agar aseptically and was incubate at room temperature for 72 hours. The various fungal isolates from each of the samples were sub-cultured to obtain pure cultures for identification. The structural features of colony, colour, extent of growth, presence or absence of mycelia, spores and the nature of colony surface were observed. Microscopic examination involved slide mounts of each isolates and stained with Lacto phenol cotton blue. Fungal identification was confirmed with

the aid of books by BARNETT & HUNTER (1999), ALEXOPOULOS & al. (2002) and AGRIOS (2005).

Antigen coated plate-enzyme-linked immunosorbent assay [ACP-ELISA] for viruses' assay

Leaf samples collected were stored at 4 °C and were tested using ACP-ELISA for the presence of Cowpea Aphid-borne mosaic virus (CabM), Black eye cowpea mosaic virus (BICMV), Cucumber mosaic virus (CM), Soybean mosaic virus (SBMV), Cowpea mottle virus (CpMov), Cowpea yellow mosaic virus (CYMV) and Cowpea mild mottle virus (CPMMV) using homologous rabbit polyclonal antiserum available in the virology unit at IITA Ibadan, following the procedure for ACP-ELISA.

Pathogenicity of the isolated organisms on healthy soybean

All the pathogens isolated from infected soybean leaf were inoculated into healthy soybean plant to determine whether they could induce similar symptoms on re-inoculation. Fungal suspension (ranges from $10^4 – 10^6$ spore /ml) was prepared from the 8 days old culture plates of the isolated fungi. The Mycelia mass of the fungus growth culture in the Petri dishes were scooped out into a sterile conical flask, which contains 10 ml of sterile distilled water, and a drop of Tween 20 detergent (for spore dispersal) was added [TODD, 2022; KEHINDE, 2008]. Inoculated soybean seedlings were covered with a transparent polythene bag for 24 hours to maintain high humidity required for disease initiation and disease symptoms were observed for up to 15 days. Distilled water served as negative control.

Data collection and analyses

Agronomic data taken includes, days to 50% flowering, days to maturity, height at harvest (cm), lodging at harvest, shattering, number of pods/plant, number of seeds/plant, grain yield (kg/ha) and 100 seed weight (g). All the data collected were subjected to analysis of variance (ANOVA), using SAS system 9.1 edition and means values separated and compared using Duncan's Multiple Range Test (DMRT) at 5% significant level of probability.

Results

Pathogens isolated from disease soybean leaf

Eight fungi were isolated from disease soybean plant in this study, *Fusarium oxysporum*, *Culvularia* spp., *Fusarium verticilloides*, *Choanephora infundibulifera* and *Aspergillus flavus* in 2015, while the same fungi were also isolated in 2016, including, *Colletotrichum gloeosporioides*, *Lasiodiplodia theobromae* and *Pestalotia* spp.

The Percentage occurrence of pathogens isolated

The percentage occurrence of fungi isolates from disease soybean leaf were presented in Figure 1. *F. oxysporum* had the highest percentage of occurrence in both years (40.91% and 22.3%), followed by *Culvularia* spp. (26.67% and 12.63%), *F. verticilloides* (22.22% and 10.20%), *C. infundibulifera* (5.00% and 17.50%) and *A. flavus* (5.20% and 8.32%). While *C. gloeosporioides* (11.5%), *L. theobromae* (7.30%) and *Pestalotia* spp. (10.25%) were only isolated in 2016.

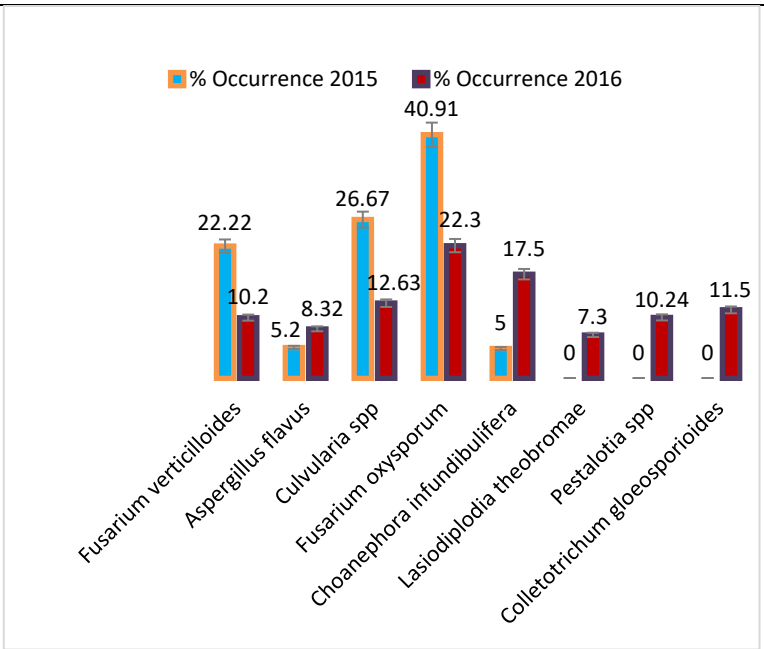

Figure 1. Percentage Occurrence of Fungi isolates from disease soybean leaf

Pathogenicity test

F. oxysporum, showed characteristics symptom of blight when used for pathogenicity on both checks, disease symptoms were first noticed on the lower (older) leaves 7 day after inoculation. The leaves turned yellow and upper leaves of infected plants appear scorched. *C. infundibulifera*, also showed blighted symptoms 7 day after inoculation with grayish patches developed on the leaves and later became necrotic.

Incidence and severity of *Fusarium* blight

Table 1 shows the average disease incidence and severity of *Fusarium* blight in early maturing lines, in 2015 and 2016 planting season, over the period of 10 weeks, after planting. TGx 1485-1D (Check) had significantly ($P \leq 0.05$) higher disease incidence of (47.90%) and (32.33%) in both years respectively, while lines TGx 1990-40F, 1989-48FN, 1989-68FN, 1990-55F and 1989-40F had significantly lower disease incidence than all other lines in 2016. The check also recorded significantly higher disease mean severities of (4.67) and (4.50) than lines TGx1989-49FN and TGx 1990-55F in 2015 and 2016 respectively.

Table 2 shows the average disease incidence and severity of *Fusarium* blight in medium maturing lines, in 2015 and 2016 planting season, over the period of 12 weeks, after planting. TGx1448-2E (Check) had significantly ($P \leq 0.05$) higher disease incidence of (47.50%) than all the lines evaluated in 2015, while the check (36.67%) and TGx 1989-42F (34.17%) had significantly higher disease incidence than other lines in 2016. TGx 1989-45F and TGx 1989-11F recorded significantly ($P \leq 0.05$) lower disease mean severity of (2.50) and (1.50) than the check (3.78) in 2015, while there was no significant difference for disease severity among all the lines evaluated in 2016.

Incidence and severity of *Choanephora* leaf blight

For early maturing lines; in 2015, TGx 1990-40F (16.17%) recorded significantly (P≤ 0.05) lower disease incidence than all the lines evaluated and the check, the same trend was also recorded in 2016 for the same line. The Check (TGx 1485-1D) had significantly (P≤ 0.05) higher mean disease severity of (4.50) and (3.33) than all the lines in both years respectively (Table 1). In the medium maturing lines, TGx1448-2E (Check) and TGx 1989-42F recorded significantly (P ≤ 0.05) higher disease incidence of (36.67%) and (34.17%) respectively than all the lines evaluated in 2015, while the check recorded higher mean severity of (2.64) and (4.36) in both years respectively (Table 2).

Table 1. Average disease Incidence and Severity for *Fusarium and Choanephora* leaf blight in early maturing soybean lines, in 2015 and 2016 planting season

Early Lines	*Fusarium* leaf blight				*Choanephora* leaf blight			
	Incidence		Severity		Incidence		Severity	
	Year's		Year's		Year's		Year's	
	2015	2016	2015	2016	2015	2016	2015	2016
TGx 1990-40F	38.50^{d}	18.90c	4.00abc	3.67bc	16.17f	13.00^{g}	3.00^{c}	2.00^{e}
TGx1989-48FN	35.90^{f}	19.50c	3.50bcd	3.50c	21.50e	16.17^{f}	3.17^{c}	2.17^{de}
TGx1989-68FN	37.33^{e}	20.20c	3.33cd	3.83bc	20.33e	16.83^{f}	3.00^{c}	2.17^{de}
TGx 1990-55F	37.50^{e}	19.00c	4.00abc	3.50c	21.17e	15.50^{fg}	3.00^{c}	2.17^{de}
TGx 1989-40F	38.50^{d}	19.83c	4.17abc	3.83bc	21.00e	16.33^{f}	3.50^{bc}	2.50^{cd}
TGx 1990-52F	36.47^{f}	30.67a	4.00abc	4.00abc	30.67bc	27.16^{cd}	3.50^{bc}	2.50^{cd}
TGx1989-49FN	34.67^{g}	31.00a	2.83d	3.67bc	29.50c	26.00^{de}	3.17^{c}	2.50^{cd}
TGx 1990-57F	43.67^{c}	26.33b	4.50ab	3.87bc	29.33c	27.67^{bcd}	3.70^{b}	2.50^{cd}
TGx 1990-46F	45.50^{b}	32.33a	4.17abc	4.00abc	26.67d	23.83^{e}	3.17^{c}	2.50^{cd}
TGx1485-1D (Check)	47.90^{a}	32.33a	4.67a	4.50a	35.50a	32.00^{a}	4.50^{a}	3.33^{a}

*Means with the same alphabet are not significantly different from each other in the same column (P ≥ 0.05)

Table 2. Average disease incidence and severity for *Fusarium* and *Choanephora* leaf blightin medium maturing soybean lines, in 2015 and 2016 planting season

Medium Lines	*Fusarium* leaf blight				*Choanephora* leaf blight			
	Incidence		Severity		Incidence		Severity	
	Year's		Year's		Year's		Year's	
	2015	2016	2015	2016	2015	2016	2015	2016
TGx 1989-45F	42.00^{f}	22.33^{cd}	2.50bc	3.60^{cde}	22.33^{cd}	24.83^{g}	2.00^{c}	3.00^{ef}
TGx 1989-11F	42.33^{e}	25.17^{c}	1.50c	3.50^{de}	25.17^{c}	25.17^{fg}	2.17^{bc}	3.33^{cde}
TGx1989-75FN	42.01^{f}	22.17^{cd}	3.56ab	3.50^{de}	22.17^{cd}	28.83^{ef}	2.00^{c}	2.83^{ef}
TGx1990-114FN	42.50^{d}	20.83^{d}	3.06ab	3.17^{e}	20.83^{d}	23.83^{g}	2.00^{c}	2.67^{f}
TGx1990-78FN	42.00^{f}	23.00^{cd}	3.50ab	3.33^{de}	23.00^{cd}	28.67^{ef}	2.22^{abc}	3.00^{ef}
TGx 1993-4FN	42.00^{f}	24.67^{c}	3.17ab	3.33^{de}	24.67^{c}	31.00^{de}	2.46^{ab}	3.33^{cde}
TGx1989-53FN	46.01^{b}	20.33^{d}	3.56ab	3.83^{cd}	20.33^{d}	25.17^{fg}	2.15^{bc}	3.17^{def}
TGx 1990-95F	44.00^{c}	22.33^{cd}	3.65ab	3.83^{cd}	22.33^{cd}	27.33^{fg}	2.17^{bc}	3.17^{def}
TGx 1989-42F	42.50^{d}	34.17^{a}	3.17ab	3.67^{cde}	34.17^{a}	36.17^{bc}	2.33^{abc}	3.56^{bcd}
TGx1990-110FN	42.00^{f}	23.37^{cd}	3.45ab	3.66^{cde}	23.37^{cd}	25.67^{fg}	2.17^{bc}	3.17^{def}
TGx1448-2E (Check)	47.50^{a}	36.67^{a}	3.78a	4.70^{a}	36.67^{a}	43.33^{a}	2.64^{a}	4.36^{a}

*Means with the same alphabet are not significantly different from each other in the same column (P ≥ 0.05)

Incidence and severity of virus diseases

Table 3 shows the average incidence and severity of virus diseases in early maturing soybean lines, in 2015 and 2016 planting season, over the period of 10 weeks after planting. TGx 1990-40F and TGx 1989-48FN recorded significantly ($P \leq 0.05$) lower disease incidence of (36.67%) and (36.83%) respectively, than TGx 1990-46F (43.37%); they also recorded significantly disease mean severity of (2.06) and (2.50) than TGx 1990-46F (3.96) in 2015. TGx 1990-40F had significantly average disease incidence of (17.34%) and also recorded lower mean disease severity than the check in 2016.

The average incidence and severity of virus diseases in medium maturing soybean lines, in 2015 and 2016 planting season, over the period of 12 weeks after planting, was presented in Table 4; TGx 1989-11F had significantly ($P \leq 0.05$) lower disease incidence of (37.83%) and mean disease severity of (1.37), than all the lines and the check in 2015. The check also recorded significantly higher disease incidence of (48.33%) than all the lines evaluated in 2016.

Virus assayed

Serological – Incidence of Soybean virus assayed in leaf sample of early and medium maturing soybean lines in 2015 and 2016 planting season were presented in Tables 3 and 4 respectively. In 2015, all early maturing lines tested negative for all the viruses assayed, except TGx 1989-68FN that tested positive for Cowpea mild mottle virus (CPMMV). All lines were positive for Cowpea mild mottle virus (CPMMV) in 2016, while the Check tested positive for Cowpea mottle virus (CpMov) in same year. In medium maturing lines, all lines were negative for all the virus assayed in 2015 and 2016, but positive for Cowpea mild mottle virus (CPMMV) in 2016.

Table 3. Average disease Incidence and Severity for Virus in early maturing soybean lines, in 2015 and 2016 planting season

Early Lines					Serology – Incidence of Soybean Virus Assayed in Leaf Sample using ACP-ELISA					
	2015		2016		2015			2016		
	Incidence	Severity	Incidence	Severity	CpMoV	CYMV	CPMMV	CpMoV	CYMV	CPMMV
TGx 1990-40F	36.67^{e}	2.06^{d}	17.34^{e}	2.97^{c}	–	–	–	–	–	+
TGx 1989-48FN	36.83^{e}	2.50^{d}	28.40^{a}	3.07^{bc}	–	–	–	–	–	+
TGx 1989-68FN	39.22^{d}	3.04^{c}	28.37^{a}	3.10^{bc}	–	–	+	–	–	+
TGx 1990-55F	39.78^{c}	3.13^{c}	22.94^{cd}	3.19^{abc}	–	–	–	–	–	+
TGx 1989-40F	38.81^{d}	3.52^{abc}	20.65^{d}	3.38^{abc}	–	–	–	–	–	+
TGx 1990-52F	38.96^{d}	3.37^{bc}	28.33^{a}	3.12^{bc}	–	–	–	–	–	+
TGx 1989-49FN	39.21^{d}	3.18^{bc}	27.00^{ab}	3.36^{abc}	–	–	–	–	–	+
TGx 1990-57F	42.22^{b}	3.76^{ab}	25.72^{abc}	3.66^{ab}	–	–	–	–	–	+
TGx 1990-46F	43.37^{a}	3.96^{a}	24.65^{bc}	3.31^{abc}	–	–	–	–	–	+
TGx 1485-1D (Check)	39.94^{c}	3.37^{bc}	28.34^{a}	3.79^{a}	–	–	–	+	–	+

Table 4. Average disease Incidence and Severity for Virus in Medium Maturing Soybean lines, in 2015 and 2016 planting season

Medium Lines					Serology – Incidence of soybean virus assayed in leaf Sample using ACP-ELISA					
	2015		2016		2015			2016		
	Incidence	Severity	Incidence	Severity	CpMoV	CYMV	CPMMV	CpMoV	CYMV	CPMMV
TGx 1989-45F	43.01^{h}	2.63^{b}	21.50^{f}	2.10^{b}	–	–	–	–	–	+
TGx 1989-11F	37.83^{k}	1.37^{c}	26.80^{bcde}	2.40^{ab}	–	–	–	–	–	+
TGx1989-75FN	42.22^{i}	2.50^{b}	24.37^{ef}	2.15^{ab}	–	–	–	–	–	+
TGx1990-114FN	44.33^{f}	2.48^{b}	30.23^{bc}	2.33^{ab}	–	–	–	–	–	+
TGx1990-78FN	41.51^{j}	2.63^{b}	25.67^{cdef}	2.11^{b}	–	–	–	–	–	+
TGx 1993-4FN	43.72^{g}	3.13^{ab}	28.73^{bcde}	2.40^{ab}	–	–	–	–	–	+
TGx1989-53FN	45.17^{c}	3.15^{ab}	24.91^{def}	2.30^{ab}	–	–	–	–	–	+
TGx 1990-95F	51.67^{a}	3.70^{a}	31.05^{b}	2.49^{a}	–	–	–	–	–	+
TGx 1989-42F	44.84^{d}	3.41^{ab}	30.53^{b}	2.19^{ab}	–	–	–	–	–	+
TGx1990-110FN	44.50^{e}	2.98^{ab}	25.50^{cdef}	2.03^{b}	–	–	–	–	–	+
TGx1448-2E (Check)	47.54^{b}	3.37^{ab}	48.33^{a}	2.29^{ab}	–	–	–	–	–	+

*Means with the same alphabet are not significantly different from each other in the same column ($P \geq 0.05$). Virus assayed by antigen coated-plate enzyme-linked immunorbent assay (ACP-ELISA); **CpMov**, Cowpea mottle virus; **CYMV**, Cowpea yellow mosaic virus; **CPMMV**, Cowpea mild mottle virus

Grain yield of soybean and resistance level to the diseases

Table 5 shows grain yield in kilogram per hectare and resistance level in early and medium maturing soybean lines, in 2015 and 2016 planting season. In early maturing lines, TGx 1989-40F (294.07 kg/ha) and TGx 1989-49FN (264.43 kg/ha) recorded significant ($P \geq 0.05$) lower grain yield than the Check (567.40 kg/ha) and TGx 1990-46F (465.97 kg/ha). The same trend was also observed for these lines in 2016. There was no significant difference in grain yield for both years in medium maturing lines.

All the lines evaluated were moderately resistant to leaf blight disease for both the maturing groups. While lines: TGx 1990-40F, TGx 1989-48FN, TGx 1989-68FN, TGx 1990-55F, TGx 1990-52F and TGx 1989-49FN are highly resistant to the virus diseases in early maturing lines; TGx 1993-4FN, TGx1989-53FN, TGx 1990-95F, TGx 1989-42F and TGx1990-110FN were moderately resistant to the same disease in medium maturing lines. The checks were moderately susceptible to all the diseases evaluated in this study in both maturity groups (Table 5).

Table 5. Grain yield and resistance level in early and medium maturing soybean lines 2015 and 2016 planting season

Early Lines	Grain yield (kg/ha)		Resistance Level		Medium Lines	Grain yield (kg/ha)		Resistance Level	
	2015	2016	*Fusarium* blight/wilt	Viral disease		2015	2016	*Fusarium* blight/wilt	Viral disease
TGx 1990-40F	345.93^{bc}	547.80^{bc}	MR	HR	TGx 1989-45F	370.40^{a}	589.70^{a}	MR	HR
TGx 1989-48FN	346.67^{bc}	563.70^{bc}	MR	HR	TGx 1989-11F	436.30^{a}	657.00^{a}	MR	HR
TGx 1989-68FN	342.20^{bc}	559.40^{bc}	MR	HR	TGx1989-75FN	288.90^{a}	509.60^{a}	MR	HR
TGx 1990-55F	402.97^{bc}	634.37^{bc}	MR	HR	TGx1990-114FN	303.70^{a}	532.00^{a}	MR	HR
TGx 1989-40F	294.07^{c}	589.83^{c}	MR	MR	TGx1990-78FN	251.90^{a}	472.70^{a}	MR	HR
TGx 1990-52F	397.03^{bc}	627.07^{bc}	MR	HR	TGx 1993-4FN	340.50^{a}	556.70^{a}	MR	MR
TGx 1989-49FN	264.43^{c}	523.00^{c}	MR	HR	TGx1989-53FN	389.60^{a}	603.40^{a}	MR	MR
TGx 1990-57F	307.43^{bc}	540.57^{bc}	MR	MR	TGx 1990-95F	403.00^{a}	621.20^{a}	MR	MR
TGx 1990-46F	465.97^{ab}	696.30^{ab}	MR	MR	TGx 1989-42F	272.60^{a}	696.80^{a}	MR	MR
TGx1485-1D (Check)	567.40^{a}	794.43^{a}	MS	MS	TGx1990-110FN	472.60^{a}	603.70^{a}	MR	MR
					TGx1448-2E (Check)	486.70^{a}	733.20^{a}	MS	MS

*Means with the same alphabet are not significantly different from each other in the same column ($P \geq 0.05$)
R= Resistant; HR= Highly Resistant; MR= Moderately Resistant; MS= Moderately Susceptible.

Discussion

Eight fungi were isolated from disease soybean plant in this study, from the pathogenicity test, only *F. oxysporum* and *C. infundibulifera* were the pathogenic ones. This result agrees with the findings of previous researchers who had associated these organisms with soybean foliar disease [FAYZALLA & al. 2009; HASHEM & al. 2009; SUBBA RAO & al. 1990]. While other fungal isolates shown no know symptom(s) when used for pathogenicity on any of the soybean checks either singly or mixed, except for *C. gloeosporioides* and *C. infundibulifera* that was re-isolated when mixed with *F. oxysporum*, they could be secondary invaders or opportunist pathogens.

F. oxysporium had the highest percentage of occurrence in both maturing lines. This pathogen can cause blight or wilt disease in soybean, and has been reported as one of the most destructive diseases of soybean [HASHEM & al. 2009; FAYZALLA & al. 2009], the pathogen can affect soybeans at any stage of development [FERRANT & CARROLL, 1981]. According to NELSON & al. (1997) and YANG (1997), *Fusarium* species are often favoured by cool temperatures, particularly in the early growing season. The decreased in moisture condition of the soil during the 2015 growing period in this study could have triggered the susceptibility of the crop to *Fusarium* blight. This result corroborates the findings of ZHANG & al. (2010) who concluded that as soil moisture becomes more limiting, soybeans become stressed, thereby increasing susceptibility to infection by *Fusarium*. DAS & al. (2019) also reported that plant infection by *Fusarium* can occur from seeds germination to mature stage, depending on the host and *Fusarium* species. *Choanephora* Leaf blight caused by fungus *C.*

infundibulifera Sacco. was also isolated in this study, as one of the foliar diseases of soybean, SUBBA RAO & al. (1990) have also documented the pathogen during their study of stem canker pathogen on soybean.

The variation in the disease incidence and severity observed in the lines evaluated, in both years could be attributed to differences in resistance status of each line and to the difference level of virulence in the pathogen. ODUBANWO & al. (2013) was also of the opinion that, soybean resistance depends on the lines level of expression and to their ability over time to tolerate the attack of the pathogens. Symptom of *fusarium* blight was more pronounced at about 6 weeks after planting, disease symptoms are first noticed on the lower (older) leaves. The leaves turn yellow and as the disease progresses, the upper leaves of infected plants wilt and appear scorched, in severe cases, the leaves dry up and drop prematurely leaving the petiole behind, which is in conformity with the report of ABIODUN & al. (2016) and NELSON & al. (1997). *C. infundibulifera* incidence was higher in 2016, there was heavy and frequent rainfall in this period, which agrees with SUBBA RAO & al. (1990) who concluded that heavy rainfall is one of the factors responsible for survival and spread of this pathogen.

Virus symptoms from this study majorly are mottling and mosaic symptoms, although other symptoms such as yellow vein banding, necrotic spots and chlorosis were also present in the field. Viruses assayed by antigen coated-plate enzyme-linked immunorbent assay (ACP-ELISA) in the laboratory were not in conformity with the field evaluation for both soybean lines in both years. This was possible because the observations in the field were based solely on visual virus-like symptoms, which were probably caused by other pathogens, physiological disorders and unidentified viruses; this finding gives credence to earlier reports of NJUKENG & al. (2013) that out of the 360 leaf samples of pepper showing virus-like symptoms collected from the field during survey followed by laboratory diagnosis using DAS-ELISA, 117 leaf samples were negative for viruses assayed. Cowpea mild mottle virus (CPMMV) is the commonest virus associated with all soybean lines used in this study, this result agrees with the conclusion of DUGJE & al. (2009) that CPMMV transmitted by whitefly (*Bemisia tabaci*) is the most prevalent virus associated with soybean mosaic disease in Nigeria.

Grain yield is considered an important indicator for any foliar disease. Grain yields among the evaluated lines varied during the two years of this study. TGx1990-46F (early maturing) and TGx1990-110FN (medium maturing) had high grain yield in both years respectively. This agrees with reports from earlier researchers who reported significant yield differences among soybean genotypes [ZHANG & ZHANG, 2000; ABLETT & al. 2000]. These lines were moderately resistant to all the diseases observed on the field, as they were able to produce high grain yield when compared with the local checks, despite the high disease incidence and severity.

Conclusion and recommendation

Leaf blight disease were the foliar diseases found to be associated with soybean lines used in this study and ecology, while Cowpea mild mottle virus (CPMMV) is the commonest virus associated with all soybean lines used. These diseases can reduce grain yield and yield traits, but it depends on the disease's severity and the genetic make-up of each soybean genotypes, these soybean lines could be utilized as parent lines for breeding against soybean foliar diseases and useful for farmers in area endemic to any of the foliar disease encounter in

this study. It is therefore recommended that further studies should be carried out on these soybean lines in other agro-ecological zone to determine the effectiveness of their resistance to foliar diseases as claimed from this study.

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