Use of Three Inoculation Methods in Screening Cowpea Genotypes for Resistance to Two *Colletotrichum* Species

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ABSTRACT

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Twelve cowpea cultivars were screened for reactions to infection by *Colletotrichum lindemuthianum* and *C. truncatum*, causal agents of anthracnose and brown blotch diseases, respectively. Three different inoculation techniques were used: spraying a spore suspension on leaves of seedlings, injecting a spore suspension into stems, and wrapping wounded seedling stems with inoculum meal. Wrapping of wounded seedlings with inoculum meal of *Colletotrichum* spp. was the best method of inoculation because it produced optimal conditions for infection and disease development. Cowpea cultivars IT82E-60, IT81D-1137, and Vita-7 were susceptible to anthracnose, whereas TVx 3236, IT81D-994, and IT81D-975 were resistant. Cultivars IT82E-60, IT82D-699, and Ife brown were susceptible to brown blotch, whereas TVx 3236, Vita-7, and IT81D-1137 were resistant.

Cowpea (*Vigna unguiculata* (L.) Walp.) is grown throughout the tropics and subtropics (9). More than 75% of the world cowpea production is in Africa, principally in Nigeria, Burkina Faso, Uganda, Niger, and Senegal (13). Cowpea anthracnose, caused by *Colletotrichum lindemuthianum* (Sacc. & Magnus) Lams.-Scrib., and brown blotch of cowpea, caused by *C. truncatum* (Schwein.) Andrus & W. D. Moore, are among the economically important diseases of cowpea in Nigeria (16).

Symptoms of anthracnose infection may appear on all above-ground parts of the cowpea plant from seedling stage to maturity, depending on the time of infection and source of inoculum (15). Leach (10) observed that infections are confined to the wall of the pod or may penetrate the endocarp and form sunken cankers 1–10 mm in diameter. At flowering, cowpea peduncles and leaves show symptoms of brown blotch infection (3). Estimated grain yield losses to anthracnose on cowpea range from 40 to 50% (19). Brown blotch has been estimated to cause up to 75% crop loss under protracted wet field conditions (3). Both diseases can reduce seed germination and seed yields (12,14).

Although chemical application is recommended for the control of anthracnose (8), the use of resistant cowpea cultivars is the most effective method of disease control. To identify sources of resistance for future breeding programs, cowpea genotypes need to be screened. We conducted the present study to develop an effective screenhouse technique for evaluating cowpea genotypes for resistance to the two diseases caused by *Colletotrichum* spp. and to determine the reactions of 12 cowpea genotypes to infection by *C. lindemuthianum* and *C. truncatum*.

MATERIALS AND METHODS

Isolates of the respective pathogens were obtained from a naturally infected cowpea plant and were cultured and subcultured separately. A spore suspension was prepared from 10- to 15-day-old subcultures of each individual fungus grown on acidified potato-dextrose agar (APDA) and was standardized to 5×10^5 spores per milliliter of distilled water with a hemacytometer. The spore suspension was used for pathogenicity studies and further inoculations as required. We prepared the inoculum meal used in this study by mixing the contents of petri dishes (i.e., APDA and 10 abundantly sporulating cultures of the respective pathogens) with 40 g of milled cowpea seeds that were ground previously into a powdery form with a Waring blender.

The experiments were performed in a screenhouse and consisted of a split-plot design with 12 cowpea cultivars as whole plots and four inoculation methods as subplots. The four inoculation methods were the control, which involved the application of sterile distilled water to seedlings with Hill Portar's hand-sprayer; the application of spore suspension of a single fungus to seedlings until run-off with Hill Portar's hand-sprayer; the injection of spore suspension (2 ml) of a single fungus on seedling stems at the area between the cotyledons and first leaves with a hypodermic syringe; and the wrapping of seedling stems with 0.5 g of the inoculum meal placed on a portion of the stem (between the cotyledons and first leaves) that was wounded after being slightly rubbed with Carborundum. The inoculum meal was wrapped up with two layers of Parafilm paper.

Four perforated plastic pots (1.2 dm^3) for each inoculation method were set up for each individual cowpea cultivar and were filled with sterilized loamy sand soil. Five seeds from each of the cultivars were sown in each pot. Each treatment combination was replicated three times during each of three trials repeatedly conducted in April, August, and November 1987. Seedlings were thinned to three at 7 days and were inoculated at 21 days after emergence, respectively. Immediately after inoculation, all seedlings were removed from the top of the screenhouse bench and placed under an enclosure made of a polyethylene sheet, which provided high relative humidity during the infection period. The seedlings were replaced on the top of the bench after 48 hr. To avoid contamination, we did the anthracnose and brown blotch experiments in different screenhouses.

On the first day that they appeared, symptoms and disease incidence were recorded based on the percentage of seedlings with symptoms of each disease. On the seventh day after inoculation, seedlings were scored for disease severity according to a modified scale (4). The scale used for anthracnose was 0, no symptoms of disease; 1, few discrete non-coalescing lesions on the leaf surface; 2, many lesions on the leaf surface occasionally coalescing; 3, coalescing lesions on the leaf surface that are continuous on more than 40 but less than 61%; 4, coalescing lesions on the leaf surface that are continuous on more than 60 but less than 81%; 5, collapse of affected part, fall of leaflet, buckling or fall of petiole, death of stem. The scale used for brown blotch was 0, no symptoms of disease; 1, up to 20% of seedling stem affected; 2, 21–40% of seedling stem affected; 3, 41–60% of seedling stem affected; 4,

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61–80% of seedling stem affected; 5, more than 80% of seedling stem affected. The reactions of the genotypes, based on the 0–5 scale, were grouped according to the following classes: 0.0–1.4, highly resistant; 1.5–2.4, moderately resistant; 2.5–3.0, moderately susceptible; and more than 3.0, highly susceptible.

Data collected during each trial were pooled and averaged over the three trials separately for each pathogen. Analysis of variance (ANOVA) as a 12 × 4 factorial (cultivar × inoculation treatments) was used, and means were separated with Duncan's multiple range test and the standard errors of the means (7).

RESULTS

Cowpea cultivars IT82E-60, IT82D-699, IT81D-994, IT84S-2246-4, and IT81D-975 were first to show symptoms of infection by *C. truncatum*; symptoms appeared within 2–3 days after seedling inoculation. Symptoms of infection by *C. lindemuthianum* first appeared on Vita-7, IT81D-1137, IT84-2246-4, IT82E-60, IT82E-16, and IT82E-32 within 2–3 days after inoculation. Other susceptible cultivars developed infection symptoms later on an irregular basis.

Cowpea cultivars IT82E-60, Vita-7, and IT81D-1137 were most susceptible to anthracnose when measured at 7 days after inoculation (Table 1). More than 40% infection resulted in severe damage of the seedlings of these cultivars. Expansion of necrotic lesions from the point of inoculation occurred, acropetally and basipetally at first, followed by radial extension along the seedling stems. Whereas infection on other cultivars ranged from 9 to 43%, no anthracnose symptoms were observed on the cultivars TVx 3236, IT81D-994, and IT81D-975.

Incidence ranging from 61 to 72% was recorded on IT82E-60, IT82D-699, Ife brown, IT81D-994, and IT84S-2246-4 after seedling inoculation with *C. truncatum* (Table 2). The severity of brown blotch and disease incidence on the cultivars followed a similar pattern (Table 2). Seedlings were toppled over at the point of inoculation when the injection and wrapping of meal methods were used; toppling was more pronounced with the latter. Acervuli were observed, sometimes scattered or clustered together on the basal stem part a few centimeters above the soil. Some of the leaves drooped, whereas others had dropped from the seedling. No brown blotch symptoms were observed on the cultivars TVx 3236, Vita-7, and IT81D-1137.

Susceptible cowpea cultivars that were inoculated by wrapping the wounded seedling stems with inoculum meal showed symptoms of disease caused by both pathogens earlier than those that were inoculated by the other techniques. The highest disease incidence after infection by the *Colletotrichum* species was recorded when wounded seedlings were inoculated by the wrapped inoculum meal method (Table 3). Similarly, individual disease severity was greatest on seedlings inoculated by the wrapping technique.

Considering the main effect of the inoculation method on the incidence of each pathogen on cowpea, we found that disease symptoms with method decreased in the following order: the wrapping of seedling stems with inoculum meal, the application of spore suspension to seedling stems with a sprayer, the injection of spore suspension on seedling stems with a syringe, and the control. The severity of brown blotch disease on the inoculated seedlings decreased in the order: the wrapping of seedling stems with inoculum meal, the injection of spore suspension on seedling stems with a syringe, the application of spore suspension to seedling stems, and the control. Severity of anthracnose on the seedlings decreased in the order: the wrapping of seedling stems with inoculum meal, the application of spore suspension to seedling stems, the injection of spore suspension on seedling stems with a syringe, and the control.

The cultivar × inoculation method interaction was significant, indicating that the reaction of the cowpea cultivars to

Table 1. Incidence and severity of anthracnose and susceptibility class and symptom appearance of 12 cowpea cultivars infected with *Colletotrichum lindemuthianum* 21 days after inoculation

Cowpea cultivar	Incidence[x] (%)	Severity[y]	Susceptibility class	Symptom appearance (days after inoculation)
TVx 3236	0 a[z]	0.0 a	Highly resistant	0
IT81D-994	0 a	0.0 a	Highly resistant	0
IT81D-975	0 a	0.0 a	Highly resistant	0
IT84S-2246-4	26 cd	1.7 b	Moderately resistant	3
IT82D-669	9 b	1.8 bc	Moderately resistant	5
Ife brown	20 c	2.0 cd	Moderately resistant	5
IT84E-124	33 e	2.1 cd	Moderately resistant	6
IT82E-32	43 f	2.3 d	Moderately resistant	3
IT82E-16	30 de	2.3 d	Moderately resistant	3
Vita-7	45 f	2.9 e	Moderately susceptible	2
IT81D-1137	45 f	3.0 e	Moderately susceptible	2
IT82E-60	58 g	3.5 f	Highly susceptible	2
Overall mean	26	2.05		
CV (%)	14	11.02		

[x] Average of three replicates and four inoculation methods.
[y] Based on 0–5 rating scale, in which 0, no symptoms of disease; 1, few discrete noncoalescing lesions on the leaf surface; 2, many lesions on the leaf surface occasionally coalescing; 3, coalescing lesions on the leaf surface that are continuous on more than 40 but less than 61%; 4, coalescing lesions on the leaf surface that are continuous on more than 60 but less than 81%; and 5, collapse of affected part, fall of leaflet, buckling or fall of petiole, death of stem.
[z] Means followed by the same letter in a column are not significantly different at $P \leq 0.05$ according to Duncan's multiple range test.

Table 2. Incidence and severity of brown blotch and susceptibility class and symptom appearance of 12 cowpea cultivars infected with *Colletotrichum truncatum* 21 days after inoculation

Cowpea cultivar	Incidence[x] (%)	Severity[y]	Susceptibility class	Symptom appearance (days after inoculation)
TVx 3236	0 a[z]	0.0 a	Highly resistant	0
Vita-7	0 a	0.0 a	Highly resistant	0
IT81D-1137	0 a	0.0 a	Highly resistant	0
IT82E-16	6 b	1.8 bc	Moderately resistant	5
IT84E-124	11 b	1.8 bc	Moderately resistant	5
IT82E-32	19 c	1.4 b	Moderately susceptible	3
IT81D-975	59 d	2.8 e	Moderately susceptible	2
IT84S-2246-4	61 de	2.6 de	Moderately susceptible	3
IT81D-994	64 def	2.1 cd	Moderately resistant	3
Ife brown	66 ef	3.1 ef	Highly susceptible	4
IT82D-699	70 fg	3.7 f	Highly susceptible	3
IT82E-60	72 g	3.9 f	Highly susceptible	2
Overall mean	35	2.19		
CV (%)	10	17.39		

[x] Average of three replicates and four inoculation methods.
[y] Based on 0–5 rating scale, in which 0, no symptoms of disease; 1, up to 20% seedling stem affected; 2, 21–40% of seedling stem affected; 3, 41–60% of seedling stem affected; 4, 61–80% of seedling stem affected; and 5, more than 80% of seedling stem affected.
[z] Means followed by the same letter in a column are not significantly different at $P \leq 0.05$ according to Duncan's multiple range test.

pathogen was different with the inoculation methods tested. Among the susceptible cultivars, the lowest values of infection (2.3–20.2% for anthracnose and 1.7–45.3% for brown blotch) were recorded on control plants, whereas infection values for each of these cultivars were not consistently significant among the other three methods of inoculation (Table 4). Whereas the application of spore suspension on seedlings of IT81D-1137, IT82E-32, IT82D-699, and Ife brown showed marked significant difference from the injection of spore suspension on the seedlings with a syringe, the reaction of IT82E-60 to infection by *C. lindemuthianum* showed no statistical difference between the two methods.

A similar trend was observed with the severity of the respective pathogens on cowpeas (Table 5). *C. lindemuthianum* was more severe on Ife brown and IT82E-32 when they were inoculated by the injection method than by the application of the spore suspension with a hand-sprayer. This was not true for most of the other susceptible cultivars. *C. truncatum* was more severe on IT82D-699 with the use of the hand-sprayer than with the use of the injection method. However, it was less severe on other cultivars when the former inoculation method was used.

In the control treatments, some cultivars showed infection even though the effect of the pathogens on these cultivars was not severe (except for IT82E-60 for each fungus and IT81D-975 for *C. truncatum*).

DISCUSSION

Discrete veinal necrosis of inoculated leaves appeared on susceptible cowpea cultivars within 2 days of inoculation. An earlier report on soybean in 1985 (11) corroborates this observation. Some cowpea cultivars, IT82E-60, IT8ID-1137, and Vita-7, were susceptible to anthracnose. The cultivars IT82E-60, IT82D-699, and Ife brown were susceptible to brown blotch and expressed symptoms of infection earlier than TVx 3236, IT81D-994, and IT81D-975, which were highly resistant to anthracnose. The cultivars TVx 3236, Vita-7, and IT81D-1137 were resistant to brown blotch and had a higher incidence of disease and the most severe symptoms of infection by the respective pathogens. Therefore, TVx 3236, the most resistant to both diseases, could be used in subsequent breeding for resistance to other diseases of cowpea. But IT82E-60, which was susceptible to both and lost its high-yielding capacity after frequent attacks from the diseases, should be recalled from circulation and placed in the gene bank. That symptoms of infection were expressed on the susceptible cultivars within 2–3 days after inoculation may indicate that the buildup of inoculum was more rapid within the seedlings of the apparently more suscep-

Table 3. Effect of inoculation methods on percentage of incidence and severity of anthracnose and brown blotch on cowpea

Inoculation method[w]	Anthracnose		Brown blotch	
	Incidence (%)	Severity[x]	Incidence (%)	Severity[y]
SS	34 ± 4 b[z]	2.06 ± 0.16 b	40 ± 6 b	2.01 ± 0.43 c
SI	20 ± 3 c	1.89 ± 0.12 b	32 ± 5 c	2.31 ± 0.36 b
MW	43 ± 5 a	2.94 ± 0.24 a	49 ± 6 a	3.17 ± 0.44 a
SC	7 ± 1 d	1.25 ± 0.08 c	21 ± 4 d	1.28 ± 0.16 d
CV (%)	25	26.67	16	32.54

[w] SS, spraying of spore suspension; SI, injection of spore suspension with hypodermic syringe; MW, wrapping of wounded seedling stems with inoculum meal; and SC, spraying of sterile deionized distilled water as the control.
[x] 0–5 Rating scale, in which 0, no symptoms of disease and 5, collapse of affected part, fall of leaflet, buckling or fall of petiole, death of stem.
[y] 0–5 Rating scale, in which 0, no symptoms of disease and 5, more than 80% of seedling stem affected.
[z] Means followed by the same letter in a column are not significantly different at $P \leq 0.05$ according to Duncan's multiple range test.

Table 4. Effect of cowpea cultivar × inoculation method interaction on the incidence of *Colletotrichum lindemuthianum* and *C. truncatum* on cowpea

Cowpea cultivar	Incidence of *C. lindemuthianum* (%)				Incidence of *C. truncatum* (%)			
	SS[z]	SI	MW	SC	SS	SI	MW	SC
IT81D-975	0.0	0.0	0.0	0.0	48.6	67.9	80.2	38.1
IT81D-994	0.0	0.0	0.0	0.0	55.7	68.5	85.7	44.0
IT81D-1137	61.7	38.0	69.0	12.7	0.0	0.0	0.0	0.0
IT82D-699	19.7	1.7	13.0	0.0	63.3	82.2	89.88	44.1
IT82E-16	35.3	19.7	56.3	7.7	0.0	3.3	19.3	0.0
IT82E-32	53.3	35.3	74.0	11.0	14.3	20.0	38.0	1.7
IT82E-60	68.3	58.0	86.7	20.3	70.6	84.5	92.7	41.3
IT84S-2246-4	34.7	17.7	46.7	3.0	60.5	66.1	83.6	32.0
Vita-7	54.0	37.7	74.3	12.7	0.0	0.0	0.0	0.0
TVx 3236	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Ife brown	31.7	10.7	36.0	2.3	62.5	75.7	80.7	45.3
IT84E-124	46.0	20.7	54.7	9.0	6.0	11.7	23.0	0.0

[z] SS, spraying of spore suspension; SI, injection of spore suspension with hypodermic syringe; MW, wrapping of wounded seedling stems with inoculum meal; and SC, spraying of sterile deionized distilled water as the control. LSD 0.05 for same method is 11.03 for *C. lindemuthianum* and 10.18 for *C. truncatum*; LSD 0.05 for same cultivar is 10.35 for *C. lindemuthianum* and 9.42 for *C. truncatum*.

Table 5. Effect of cowpea cultivar × inoculation method interaction on the severity of *Colletotrichum lindemuthianum* and *C. truncatum* on cowpea

Cowpea cultivar	Severity of *C. lindemuthianum*[x]				Severity of *C. truncatum*[y]			
	SS[z]	SI	MW	SC	SS	SI	MW	SC
IT81D-975	1.0	1.0	1.0	1.0	4.5	3.5	5.0	2.5
IT81D-994	1.0	1.0	1.0	1.0	1.0	3.0	2.3	1.0
IT81D-1137	3.3	2.7	4.7	1.3	1.0	1.3	1.5	1.5
IT82D-699	2.0	1.7	2.3	1.0	5.0	1.3	4.3	1.0
IT82E-16	2.3	2.0	2.3	1.0	1.0	2.0	3.3	1.0
IT82E-32	2.0	2.3	3.7	1.0	1.0	1.0	1.0	1.0
IT82E-60	3.3	2.3	5.0	3.3	3.3	4.3	4.8	2.4
IT84S-2246-4	1.7	1.7	2.3	1.0	2.5	3.0	4.0	1.0
Vita-7	2.7	2.3	4.7	2.0	1.0	1.0	1.3	1.0
TVx 3236	1.0	1.1	1.0	1.0	1.0	2.3	4.3	1.0
Ife brown	1.7	2.7	2.7	1.0	1.0	4.3	4.8	1.0
IT84E-124	2.3	2.0	3.0	1.0	1.8	1.0	1.8	1.0

[x] 0–5 Rating scale, in which 0, no symptoms of disease and 5, collapse of affected part, fall of leaflet, buckling or fall of petiole, death of stem.
[y] 0–5 Rating scale, in which 0, no symptoms of disease and 5, more than 80% of seedling stem affected.
[z] SS, spraying of spore suspension; SI, injection of spore suspension with hypodermic syringe; MW, wrapping of wounded seedling stems with inoculum meal; and SC, spraying of sterile deionized distilled water as the control. LSD 0.05 for same method is 0.86 for *C. lindemuthianum* and 1.03 for *C. truncatum*; LSD 0.05 for same cultivar is 0.89 for *C. lindemuthianum* and 1.00 for *C. truncatum*.

tible cultivars than within others that were less susceptible. Both species of *Colletotrichum* are seed-transmitted (3,5,6). Therefore, it is possible that there were differentials in inoculum carryover by the seeds of the different cultivars. This, in addition to the standardized inoculum used, might have enhanced symptom manifestation on the cowpeas.

With the injection method, the initial inoculum injected into seedlings might not be enough to cause infection in the susceptible cultivar because of restriction of movement. Seedlings inoculated by the wrapping of wounded stems with inoculum meal showed the most severe symptoms after infection by the two *Colletotrichum* species in this study. For Phytophthora root rot infection of *Banksia* species, a similar report has been made (2). However, this method of inoculation does not actually simulate the natural field situations in which the inoculum is in constant contact with the plant.

As with the injection method, inefficient deposition of the inoculum and restriction of the movement of the same inoculum within the tissue could occur with the wrapping method. The method cannot, therefore, be recommended because it may overcome certain types of resistance. Moreover, it is more tedious and time-consuming than the method of spore suspension by spraying. Again, if a large number of propagules of a fungus are deposited in the tissue, close to each other, crowding and the attendant negative interaction among the conidia (competition for space, germination, growth, development, and colonization of the pathogen within the host tissue) may occur (20). Also, antagonism among propagules may arise, thus reducing the effective inoculum density in the tissue of the plant in the field. This reduction may delay the development of symptoms. Hence, a plant breeder who has thousands of plants to inoculate would probably not be able to use this method. Alternatively, the breeder could spray the plants with a spore suspension to reduce the amount of work and the length of time required. Tu and Aylesworth (18) demonstrated that spraying inoculum suspension leads to variable infection because of nonuniform distribution of inoculum during spraying.

In our studies, certain cultivars showed symptoms of infection even though they were not inoculated. According to Tiffany (17), this could be attributed to the successful establishment of the mycelium from the previous season within the seed from the field. Thus, the possibilities of the symptoms developing from seed infection from the previous season may account for development of disease in control plants. Similar results were obtained in earlier studies with *C. lindemuthianum* (1)

LITERATURE CITED

1. Dhingra, O. D., Fernandez, C. M. A., and Kushalappa, A. C. 1986. Lack of relationship between field incidence of bean anthracnose and production of seeds transmitting *Colletotrichum lindemuthianum*. Fitopatol. Bras. 11:95-101.
2. Dixon, K., Thinlay, W., and Sivasithamparam, K. 1984. Technique for rapid assessment of tolerance of *Banksia* spp. to root rot caused by *Phytophythora cinnamomi*. Plant Dis. 68:1077-1080.
3. Emechebe, A. M. 1981. Brown blotch of cowpea in Northern Nigeria. Samaru J. Agric. Res. 1:20-26.
4. Emechebe, A. M. 1985. Screening advanced cowpea breeding lines for resistance to anthracnose and brown blotch at Ibadan. Pages 90-100 in: Grain Legume Improvement Programme, Annual Report for 1985. International Institute of Tropical Agriculture, Ibadan, Nigeria.
5. Emechebe, A. M., and McDonald, D. 1979. Seed-borne pathogenic fungi and bacteria of cowpea in Northern Nigeria. Pest Artic. News Summ. 25:401-404.
6. Esuruoso, O. F. 1975. Seed-borne fungi of cowpea in Western Nigeria. Niger. J. Plant Prot. 1:87-90.
7. Gomez, K. A., and Gomez, A. A. 1984. Statistical Procedures for Agricultural Research. 2nd ed. John Wiley and Sons, New York. 680 pp.
8. International Institute of Tropical Agriculture. 1974. Annual Report for 1973. Ibadan, Nigeria. 199 pp.
9. Johnson, D. T. 1970. The cowpea in the Africa areas of Rhodesia. Rhod. Agric. J. 67:61-64.
10. Leach, J. G. 1923. The parasitism of *Colletotrichum lindemuthianum*. Tech. Bull. Minn. Agric. Exp. Stn. 14. 41 pp.
11. Manandhar, K. K., Indra, T., Singh, S. R., Hartman, G. L., and Sinclair, J. B. 1985. Penetration and infection of soybean leaf tissues by *Colletotrichum truncatum* and *Glomerella glycines*. Fitopathol. Bras. 9:365.
12. Onesirosan, P. T., and Barker, L. N. 1971. Stem anthracnose of cowpea in Nigeria. Plant Dis. Rep. 55:820.
13. Rachie, K. O., and Rawal, K. M. 1976. Integrated approaches to improving cowpeas, *Vigna unguiculata* (L.) Walp. IITA Tech. Bull. 5. 36 pp.
14. Saxena, R. M. 1984. Evaluation of injection percentage and crop estimates of some seed borne infection of green and black gram in Utah Pradesh. Indian J. Plant Pathol. 2:146-148.
15. Schwartz, H. F., and Galvez, G. E. 1980. Bean Production Problems: Disease, Insect, Soil and Climatic Constraints of *Phaseolus vulgaris*. Centro Internacional de Agricultura Tropicale (CIAT). 424 pp.
16. Singh, S. R., and Allen, D. J. 1979. Cowpea Pests and Diseases. International Institute of Tropical Agriculture, Ibadan. Nigeria. Manual Ser. 2. 113 pp.
17. Tiffany, L. H. 1951. Delayed sporulation of *Colletotrichum* on soybean. Phytopathology 41:975-985.
18. Tu, J. C., and Aylesworth, J. W. 1980. An effective method of screening white (Pea) bean seedlings (*Phaseolus vulgaris* L.) for resistance to *Colletotrichum lindemuthianum*. Phytopathol. Z. 99:131-137.
19. William, R. J. 1975. Diseases of cowpea (*Vigna unguiculata* (L.) Walp.) in Nigeria. Pest Artic. News Summ. 21:263-267.
20. Wood, R. K. S. 1989. Physiological Plant Pathology. 2nd ed. Blackwell Scientific Publications, Oxford. 497 pp.