THE BIOLOGY AND CONTROL OF *CONIELLA* LEAF SPOT OF KENAF, *HIBISCUS CANNABINUS* L., INDUCED BY *CONIELLA MUSAIAENSIS* B. SUTTON VAR *HIBISCI* B. SUTTON

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DECLARATION

I hereby declare that this thesis has been written by me and that it is a record of my own research work. It has not been presented before in any previous application for higher degree.

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CERTIFICATION

This thesis entitled "THE BIOLOGY AND CONTROL OF CONIELLA LEAF SPOT OF KENAF, HIBISCUS CANNABINUS L., INDUCED BY CONIELLA MUSAIAENSIS B. SUTTON VAR. HIBISCI B. SUTTON" by Adepoju Aremu Adeoti meets the regulations governing the award of the degree of DOCTOR OF PHILOSOPHY of Ahmadu Bello University, and it approved for its contribution to knowledge and literary presentation.

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DEDICATION

This thesis is dedicated to my wife (Funke) and our children (Abidemi, Adenle, Adewale, Adepeju, Adeyinka, Adekunle and Adedotun) in loving appreciation of their encouragements and sacrifice throughout the period of this work.

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ABSTRACT

Symptomatological studies and pathogenicity tests confirmed that the Coniella leaf spot of kenaf observed in Mokwa in Nigeria was induced by Coniella musaiaensis B. Sutton var hibisci B.Sutton (IMI NO. 280998 and 2809996). Field survey conducted in the southern Guinea savanna of Nigeria in 1984 indicated that C. musaiaensis induced between 41,5 and 98.2 percent damage to the leaves. Out of 57 kenaf cultivars screened on the field and glasshouse for resistance to the pathogen, only Tianung No.lwas free of the disease, three other cultivars, AMC 108 kenaf, A60-282 and local line 1 exhibited high resistance.

The month of May at Mokwa, in the southern Guinea savanna, was the best for planting kenaf to avoid severe disease attack and to obtain high grain and fibre yields Planting as from July resulted in flowering in August(when the day length has fallen), severe attack by the leaf spot disease and low grain and fibre yields.

Application of NPK fertilizer increased kenaf growth and fibre yield but increased Coniella leaf spot severity on the crop. Supply of N as sole fertilizer also predisposed kenaf plants to attack by the **leaf** spot. In both cases, the age of plant **at first** apprearance of disease symptom was reduced significantly (P = 0.05) when compared with plots where NP and NK fertilizers were used. Supply of NP or NK fertilizers significantly (P = 0.05) delayed the time of disease attack on kenaf, reduced disease severity and increased both grain and fibre yields.

In-vitro assessment of four fungicides showed that carbendazim andbenemy1 completely inhibited the mycelial growth of the fungus in agar medium at the commercial dosage and at lower concentrations tested. Calixin M and Mancozeb inhibited mycelial growth in culture only at double the commercial dosage. The four fungicides significantly (P = 0.05) reduced the disease severity when sprayed at two-weekly interval compared with the check that was sprayed with water. There was no significant difference (P = 0.05) in disease severity between kenaf plants sprayed with carbendazim at 7-day and 14-day intervals. There were also no significant differences (P = 0.05) in disease severity when the four fungicides were sprayed at 21—and 28-day intervals when compared with the controls in the two years. Retted fibres from fungicide-treated plots showed that there was no need to spray for Coniella leaf spot control if the ultimate aim of planting kenaf was fibre production, because the check plots gave fibre yields comparable in weight with the sprayed plots. However, it is necessary to spray the plots if grain production is required since the fungicide-treated plots gave significantly (P = 0.05) higher grain yield than the unsprayed plots.

Out of the 17 plant species belonging to 9 taxonomic families tested for susceptibility to C. musaiaensis, five species - okra, Abelmeschus esculoentus L.; cotton, Gessypium hirsitum L.; pepper, Capsicum annun L.; and two cultivars of reselle (Hibiscus sabdariffa) were found to be susceptible. H. sabdariffa has been reported earlier as an alternative host of the pathogen while the other plants are being reported for the first time.

Based on the interaction in the glasshouse between 9 selected kenaf cultivars (3 resistant, 3 moderately resistant and 3 susceptible) as host differentials and 51 isolates of C. musaiaensis collected from many kenaf growing areas in the southern and northern Guinea savanna zones of Nigeria, six pathogenio groups of the organism were proposed. The immediate implication of this is is that more races of the pathogen are probably existing in Nigeria.

TABLE OF CONTENTS

		PAGE
TITLE PAGE		i
DECLARATION		ii
CERTIFICATION		iii
DEDICATION		iv
ACKNOWLEDGEMENTS		v
ABSTRACT		vii
TABLE OF CONTENTS		x
LIST OF TABLES		xiv
LIST OF FIGURES		xvi
LIST OF PLATES		xvii
LIST OF APPENDICES		xviii
CHAPTER:		
1	INTRODUCTION	1
2	LITERATURE REVIEW	5
3	MATERIALS AND METHODS	8
3.1	Experimental Location	8
3.2	Materials	8
3.3	Methods	8
3.3.1	Sterilisation of media and glasshouse	8
3.3.2	Experimental design	9
3.3.3	Design of instrument for measuring the height and butt diameter of kenaf	9
3.3.4	Retting kenaf for fibre	10
4	PRELIMINARY STUDIES	14
4.1	Materials and Methods	14

Capter		PAGE
4.1.1	Symptomatology	14
4.1.2	Isolation and identification of the pathogen	14
4.1.3	Pathogenicity test	16
4.2	Results and Discussion	18
4.2.1	Symptomatology	18
4.2.2	Isolation and identification of the pathogen	21
4.2.3	Pathogenicity test	24
5	SURVEY OF THE INCIDENCE AND SEVERITY OF CONIELLA LEAF SPOT OF KENAF	25
5.1	Materials and Methods	25
5.1.1	Area surveyed	25
5.1.2	Survey methods	25
5.1.3	Disease assessment	27
5.2	Results and Discussion	27
6	SCREENING KENAF CULTIVARS FOR RESISTANCE TO CONIELLA LEAF SPOT	31
6.1	Materials and Methods	31
6.1.1	Screening under field condition	31
6.1.2	Screening in the glasshouse	32
6.2	Results and Discussion	33
7	EVALUATION OF THE EFFECT OF SOME CULTURAL PRACTICES ON THE INCIDENCE AND SEVERITY OF CONIELLA LEAF SPOT OF KENAF	44
7.1	Effect of date of planting on the development of Coniella leaf spot of kenaf	44
7.1.1	Introduction	44
7.1.2	Materials and Methods	45
7.1.3	Results and Discussion	46

Chapter		PAGE
7.1.3.1	Correlation Analysis	49
7.2	Effect of N, P and K fertilizers on the development of Coniella leaf spot	54
7.2.1	Introduction	54
7.2.2	Materials and Methods	55
7.2.3	Results and Discussion	57
8	CHEMICAL CONTROL OF CONIELLA LEAF SPOT OF KENAF	63
8.1	Introduction	63
8.2	Materials and Methods	63
8.2.1	In-vitro assessment of the effects of dosage of 4 fungicides on growth and conidial production in C.musaiaensis.	63
8.2.2	Effects of fortnightly sprays of four fungicides on Coniella leaf spot of kenaf.	66
8.2.3	Effect of spraying intervals of four fungicides on the Coniella leaf spot and yield of kenaf	67
8.3	Results and Discussion	68
8.3.1	In-vitro assessment of four fungicides for their efficacies against Coniella musaiaensis	68
8.3.2	Effect of fortnightly sprays of four fungicides on Coniella leaf spot of kenaf	74
8.3.3	Effect of spraying intervals of four fungicides on Coniella leaf spot and yield of kenaf at Mokwa in 1984 and 1986	76
9	DETERMINATION OF ALTERNATIVE HOSTS OF CONIELLA MUSAIAENSIS	84
9.1	Introduction	84
9.2	Materials and Methods	84
9.3	Results and Discussion	85

Chapter		PAGE
10	PATHOGENIC VARIATION AMONG ISOLATES OF CONIELLA MUSAIAENSIS	90
10.1	Introduction	90
10.2	Materials and Methods	90
10.2.1	Selection of host differentials	90
10.2.2	Collection of diseased specimens	91
10.2.3	Isolation of the pathogen and preparation of inoculum	95
10.2.4	Inoculation of host differentials	96
10.3	Results and Discussion	97
	SUMMARY AND CONCLUSION	107
	LITERATURE CITED	113
	APPENDICES	118

LIST OF TABLES

TABLE		PAGE
1	Incidence and severity of Coniella leaf spot of kenaf on selected farms in parts of Niger and Kaduna States in 1984.	29
2	Field and glasshouse reactions of kenaf cultivars to inoculation with Coniella musaiaensis in 1984, 1986 and 1988.	34
3	Kenaf cultivars that exhibited consistent reactions to field inoculation with C. musaiaensis in 1984 and 1986.	38
4	Kenaf varieties that exhibited inconsistent reactions to field inoculation with C. musaiaensis in 1984 and 1986.	40
5	Effect of date of planting on Coniella leaf spot and yield of kenaf cultivar Cuba 108 in Mokwa - 1984.	47
6	Effect of date of planting on Coniella leaf spot and yield of kenaf cultivar Cuba 108 in Mokwa - 1986.	48
7	Correlation between Coniella leaf spot severity score and kenaf yield components at Mokwa, 1984.	50
8	Correlation between Coniella leaf spot severely score and kenaf yield components at Mokwa, 1986.	51
9	Effect of M, P and K fertilizers on the development of Coniella leaf spot disease of kenaf in Mokwa, 1984.	58
10	Effect of M, P and K fertilizers on the yield and Coniella leaf spot disease development on kenaf in Mokwa, 1986.	59
11	Chemical nature, trade and common names of fungicides evaluated in the laboratory and field for their efficacies against C. musaiaensis.	64
12	Effect of concentrations of Delsene - M, Dithane M-45, Calixin M and Benlate on the growth of C.musaiaensis in agar medium.	69

13	Screening four fungicides for their efficacies in controlling the leaf spot of kenaf in Mokwa, 1983	75
14	Effect of spraying intervals of four fungicides on the severity of Coniella leaf spot and yield components of kenaf in Mokwa, 1984	77
15	Effect of spraying intervals of four fungicides on the severity of Coniella leaf spot and yield components of kenaf in Mokwa, 1986.	79
16	Pathogenicity of C. musaiaensis in some species of crops and weeds in the glasshouse at Samaru.	86
17	Sources of C.musaiaensis isolates used in the study of pathogenic variation in C.musaiaensis.	92
18	Interaction between isolates of C. musaiaensis and 9 cultivars of kenaf in the glasshouse.	98
19	Pathogenic groups of isolates of C.musaiaensis on 9 kenaf cultivars in the glasshouse.	102

LIST OF FIGURES

FIGURE		PAGE
1	Map of Nigeria showing the savanna ecological zones and the area surveyed for Coniella leaf spot of kenaf induced by C.musaiaensis.	26

LIST OF PLATES

PLATE		PAGE
1	Instrument for measuring the height of kenaf plants.	11
2	Instrument for measuring the butt diameter of kenaf plants.	12
3	Typical lesions of Coniella leaf spot of kenaf.	19
4	Stem dis-back due to severe infection of kenaf stem by C.musaiaensis var hibisci.	20
5	Culture of C. musaiaensis var hibisci formed in plates incubated in daily alternating cycles of light and darkness	22
6	Uniformly brown colony of C. musaiaensis var hibisci produced in plate incubated under continuous darkness.	23
7	Effect of concentration of carbendazim on the mycelial growth of C. musaiaensis in potato dextrose agar medium.	70
8	Effect of concentration of beromyl on mycelial growth of C. musaiaensis in PDA.medium.	71
9	Effect of concentration of mancozeb on the mycelial growth of C. musaiaensis in PDA medium.	72
10	Effect of concentration of Calixin M on the mycelial growth of C. musaiaensis in PDA medium.	73
11		

LIST OF APPENDICES

APPENDIX		PAGE
1	Identification Services: Commonwealth Mycological Institute, Surrey, England.	118
2	Selected properties of the soil at the Experiemental site in Mokwa - 1984 and 1986,	119
3	Mean Temperature and Rainfall at Mokwa - 1984 and 1986.	120
4	Interaction between isolates of Coniella musaiaensis and selected cultivars of kenaf in the glasshouse.	121
5	Brief description of Coniella species.	123
6	Brief description of kenaf, Hibiscus cannabinus L.	124

CHAPTER 1

INTRODUCTION

Kenaf, Hibiscus cannabinus L., is a source of soft fibre. Its origin is not quite known, but Delziel (1948) reported that it could be found in open savanna country between Senegal and Nigeria. He suggested that Angola was its centre of diversity and probably its centre of origin. Dempsey (1975) also asserted that kenaf originated from Africa. He estimated that kenaf has been grown for thousands of years in Africa for fibre and food. Among the countries where kenaf is grown are India, China, Russia (USSR), Iran, Mozambique, Taiwan, El-Salvador, Benin Republic, Cote d'Ivoire and Nigeria. Guest (1979) identified India and Bangladesh as the major producers of jute and kenaf.

The crop is cultivated in large areas all over the world. In India, for instance, as much as 27,000 hectres are cultivated in Andhra Pradesh alone (Kundu, 1964). Dempsey (1975) reported that more than 40,000 hectres were under cultivation with kenaf in USSR and that by 1959 kenaf production had reached 210,000 metric tons in China. Although kenaf is found growing in many parts of Nigeria, there is no accurate record of the extent of its cultivation.

The rate of cultivation of kenaf attests to the usefulness of the crop. It is remarkably versatile as a multi-use crop. There is scarcely any part of the plant that is wasted.

For instance, observations by the author showed that the leaves are usually harvested green and eaten as vegetable in many parts of Niger and Kaduna States of Nigeria. The leaves are also used as silage. Wing (1967) reported that kenaf silage is acceptable to cattle, while Killinger (1965) indicated that kenaf foliage contained from 25 to more than 30 percent crude protein.

The fibre obtained from kenaf also has various uses. Kirby (1963) reported that as much as 40 to 60 percent of kenaf fibre is mixed with jute for making hessian sacks. He also reported that in Bombay Province of India, most of the fibre is used locally for rope making, in strings, net for carrying cotton, carpets, mats and chair backs.

In Nigeria, the fibre produced by farmers is used mostly for rope making. The stripped woody stem of kenaf is generally used for fuel; sometimes it is used in erecting small fencing around the farmer's house and chicken pens (Dempsey, 1975). However, some researchers (e.g. Nieschlag et al., 1960; Nelson et al., 1961; Clark and Wolff, 1962; Annon, 1978) have reported that kenaf stem is a good source of pulp for the paper industry.

Dempsey (1972) reported that the kenaf seed contains about 20 percent by volume of edible oil. This oil compares favourably with that obtained from cotton seed. Annor (1978) also reported that oil obtained from kenaf is good for margarine and for high protein stock feed.

This very useful crop is affected by a lot of diseases, some of which include dry stem rot induced by Macrophomina phaseoli (Maubl.) Ashby; leaf spot induced by Cercospora hibisci. Tracy and Eark; leaf blight induced by Phyllosticta hibisci peck; stem rot induced by Diplodia hibiscina Cke. & Elbi. and anthracnose induced by Colletetrichum hibisci Polacci (Pursoglove, 1968).

While some work has been carried out in Nigeria on the disease problems of other soft fibres, such as roselle (H.sabdariffa), by Olunloyo and Adeniji (1976; 1980), there is little record of work done on the diseases of kenaf. However, Emechebe (1980) listed eight pathogens responsible for kenaf diseases in northern Nigeria. Among these is Coniella hibisci Morgan-Jones incitant of a leaf spot. This is probably the same fungus as C.musaiaensis var hibisci.

Field observations by the author in Niger and Kaduna States, where kenaf is grown extensively, showed that several diseases attack it; prominent among these is the Coniella leaf spot. This disease is severe in many places that kenaf is grown as from about six weeks after planting resulting in premature defoliction and sometimes death of the affected plants.

The few reports available in literature on the pathogen did not include any control measures for the disease either in Nigeria or in any other part of the world.

The objectives of this study are, therefore, to

(a) conduct field surveys in different kenaf growing areas of Nigeria, particularly in the southern Guinea savanna zone, with a view to determining the incidence and severity of the disease on kenaf plants;

(b) establish the pathogenicity of C.musaiaensis on kenaf;

(c) evaluate the effect of some cultural practices adopted by the farmers on the development and severity of the leaf spot;

(d) study pathogenic variation in C.musaiaensis;

(e) evaluate the efficacy of some fungicides in the control of the disease; and

(f) search for alternative hosts of the pathogen among the flora in the kenaf growing areas.

CHAPTER 2

LITERATURE REVIEW

One of the major constraints to kenaf production is its susceptibility to many diseases. Dempsey (1975) reported that practically all kenaf diseases are caused by various seed-and soil-borne fungi. He considered anthraenose induced by Colletotrichum sp. to be a major kenaf disease because it is widespread and very destructive.

Another major disease of kenaf is Rhizoctonia root rot induced by Rhizoctenia solani Kuhn, the imperfect stage of Thanatephorus cucumeris (Franak) Donk. Another stem disease is the eye-rot induced by Myrothecium roridum Tode ex Fr. This was first reported in India (Das and Dikshit, 1975) and could result in the breaking of the stems in severe cases.

Some leaf spot diseaseshave also been reported on kenaf. Kundu (1964) reported thatCCercospora leaf spot was the major leaf spot disease of kenaf in India, Japan and Taiwan, while Protesenko (1967) in USSR, reported a damping-off disease of kenaf induced by Alternaria sp.

As shown above, some work has been carried out on the biology and control of kenaf diseases in different parts of the world. In Nigeria, not much work has been done on kenaf diseases. However, Abdullahi (1970) first reported that pests and diseases were potentially important in the development of bast fibre industry in Nigeria. He observed that a few diseases attack

kenaf and reselle in Nigeria. Among these were leaf spot diseases common at Jema'a and Zonkwa in Kaduna state which included anthracnose induced by Colletotrichum hibisci and "leaf burn" induced by Corticium solani. Others are stem diseases such as Phytophthora blight induced by Phythophthora nicotianae which usually results in rotting of stems and roots.

The first major record of kenaf diseases in Nigeria was by Emechebe (1980) who listed eight different orgnisms responsible for causing kenaf diseases in the savanna and semi-arid zones of Nigeria. These include Phytophthora parasitica Dastum, incitant of the stem rot disease of kenaf, Corticium solani (Prill. & Delaer.) inducing the web blight and leaf spot disease. Also included were leaf spot diseases induced by Coniella hibisci Morgan-Jones, Coniella musaiaensis Sutton, and Ascochyta blight induced by Ascochyta abelmoschi Harter. Others listed were the collar rot (Sclerotium rolfsii Sacc.) vascular wilt, Fusarium oxysporum Schlecht, and stem rot induced by Rhizoctonia solani Kuehn and Macrophomina phaseolina (Tassi) Gold.

Of all the diseases listed above, Coniella leaf spot is the most destructive (Annon 1983). Its attack always starts with a water soaked spot; this expands extensively covering a large part of the leaf. At the same time, a dark brown area appears in the centre of the lesion. The brown area is usually covered by a large number of Pycnidia that give the

spot a dark brown appearance. The attack could lead to premature, complete defolination of the kenaf plant. On the average, between 75 and 80 percent of the plants were attacked when sampling was carried out in September (Annon., 1983). Loss of seed viability of up to 66 percent has also been attributed to the leaf spot (Idem, 1982 - personal communication).

The causal organism has been found to be Coniella musaiaensis B. Sutton var. hibisci B. Sutton (Khatua and Maiti, 1977). It is not certain whether the two causal agents of leaf spot disease listed by Emechebe (1980) as C.hibisci Morgan-Jones and C.musaiaensis Sutton were synonymous with C.musaiaensis var hibisci.

Apart from above identifications, it appears, from available literature, that no work has been carried out on the causal agent of the Coniella leaf spot.

Coniella species have also been recorded on other crops in Nigeria. For instance, Bailey (1966) reported Coniella diplodiella (Speg) Petr. & Syd. as inducing the leaf blotch disease of grape vine - Vitis vinifera L. - while Emechebe (1980) listed Coniella sp as inducing the leaf and stem blight of reselle, Hibiscus sabdariffa L.

CHAPTER 3

MATERIALS AND METHODS

3.1 Experimental Location

Mokwa (9^{o} 11'N; 5^{o} 04' E), in the southern Guinea savanna, was chosen for the field experiments. It has a mean annual rainfall of 1, 123 mm which is also confined to a single rainy season of 7 months (April to October). The rainfall has two peaks usually, in July and September. The soil is deep, freely drained, sandy loam, formed over cretaceous sand stone and mapped as ferralitic by D'Hoore (1964).

3.2 Materials

Most of the materials needed were the common anyltical agar and glassware obtainable in the laboratory. The chemicals used were fertilizer and fungicides. Other materials were seeds of kenaf cultivar, Cuba 108 and those of 57 other cultivars.

3.3 Methods

3.3.1 Sterilisation of media and glassware

The agar medium used throughout this study was sterilized in 250 ml Erlemeyer flask by autoclaving for 15 minutes at 1 kg/cm^2 pressure.

Glassware, mostly petri dishes, were put in steel canisters, covered and sterilized in the oven for not less than 6 hours at 120°C, to ensure adequate sterilization. The soil used for glasshouse work was heat-sterilized in an electric soil sterilizer.

3.3.2 Experimental Design

Randomized complete block design was used for all the field experiments. The number of replicates of respective treatments of various experiments was not less than three. In the field trials each plot was surrounded by a border row of one meter. The experiments were planted on the flat.

In the glasshouse studies, the plastic pots with artificially inoculated plants were always arranged in completely randomized block design.

3.3.3 Design of instruments for measuring the height and butt diameter of kenaf

As a result of difficulties encountered in measuring tall kenaf plants and, in order to facilitate quick and accurate measurements of the plant heights and but diameters respectively, two measuring instrument were designed.

For plant height measurement, two wooden planks were calibrated from 0 to 4.5 m; the first being from 0 to 2.5 m and the other from 2.5 to 4.5 m.

The pieces of plank were held together by a zinc case in such a way that it was possible for one peice of plank to slide over the other. At the top of the two pieces of plank, another one was attached at right angle. This third piece was not calibrated; it was meant for determining the top of the kenaf plant (Plate 1).

For measurement of butt diameter, an inverted L-shaped instrument was designed. The shorter length was calibrated in milimeters. Holding the longer part, the instrument was placed on the ground at the base of the stem and the diameter was read directly from the short arm (Plate 2).

3.3.4 Retting kenaf for fibre

The kenaf plant was cut at the ground level at flowering. The leaves were removed using a knife. The stems were then bound together with rope and lowered into water in a stream. Each bundle was held down the bottom of the stream by placing heavy objects, such as stone, on it.



Plate 1. Instrument for measuring the height of kenaf plants.

... Library,
... Bello University,
Zaria - Nigeria.

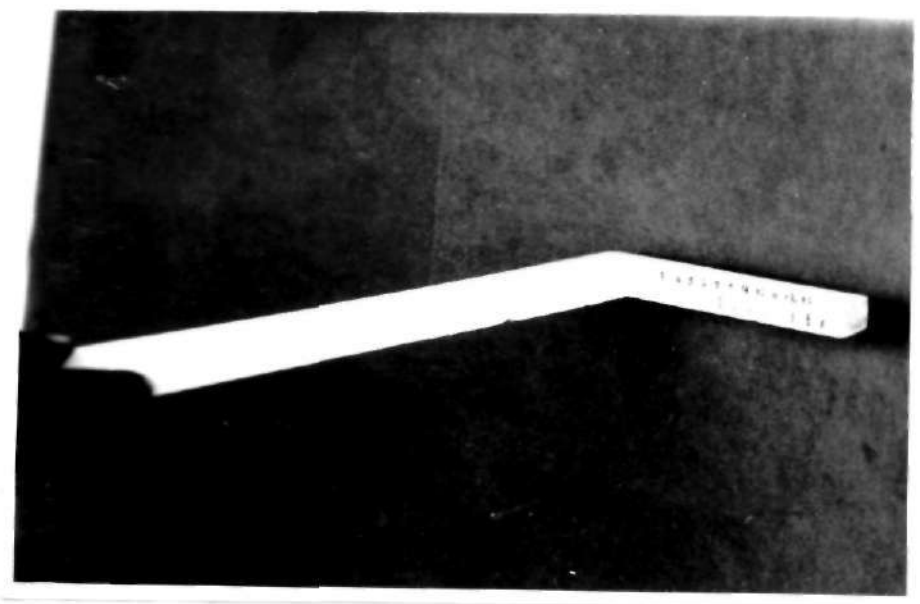


Plate 2. Instrument for measuring the butt diameter of kenaf plants.

It was left for 3 weeks to ret after which the wood was removed and the retted bark were gathered and arranged linearly. The bark was washed by squeezing several times until the fibre became clean. The fibre was dried to constant weight in the sun.

CHAPTER 4

PRELIMINARY STUDIES

4.1 Materials and Methods

4.1.1 Symptomatology

Visits were made at forthnightly intervals to homesteads and kenaf plantations in and around Mokwa as from May, when the kenaf was planted by the farmers. Observations were made on the symptoms of the disease, period of the year when the symptoms appear and the disease incidence. Infected leaves were collected in labelled paper envelopes and taken to the laboratory for the isolation of the pathogen.

4.1.2 Isolation and Identification of the pathogen

The leaf lesions were cut into small pieces (about 1 cm^2) close to the margin of the advancing lesion. This was to avoid contamination by saprophytes which might have invaded the older parts of the lesion.

The leaf pieces were surface sterilised by immersing each piece in 1% solution of sodium hypochlorite (NaOCl) for about one minute. The pieces were then washed in two changes of sterile distilled water, dried between layers of

filter paper, and aseptically plated (two per plate) on 24-hour old potato dextrose agar medium to which streptomycin (10 mg/litre) had been incorporated (PDAS). The plates were incubated at room temperature (24 - 30^{o}C) for 3 days.

After 72 hours, fungal hyphae growing out of the plant materials were transferred into fresh PDAS plates which were then incubated for 7 days before microscopic examination was carried out.

Monoconidial culture of the organism was obtained as follows. Conidia suspension was obtained by flooding 7-day old culture with 10 ml of sterile distilled water for two minutes. This was shaken continuously before the resultant conidial suspension was poured into a sterilized McCartney bottle.

With the use of sterile pipette, 1 ml of the suspension was transferrred into 9 ml of sterilized distilled water in another sterilized McCartney bottle. This was also shaken vigorously; 1 ml of the suspension was similarly transferred into 9 ml of steriled distilled water in another sterilized bottle. The serial dilutions were continued until a 1:10^{7} dilution was obtained.

Five drops (0.5ml) from each dilution were placed on a PDAS plate which was gently rotated to facilitate even distribution of the suspension.

The plates were incubated for 48 hours to allow young colonies to develop from germinated conidia. Only colonies that clearly orginated from single conidia were picked using flamed, inoculating needle from $1:10^4$ serial dilution into fresh PDAS plates and incubated for 7 days at room temperature ($24-30^{o}C$).

Colony characteristics of the organism were observed. With the use of micrometer eye piece and the light microscope, the sizes of 1,000 pycnidia and 1,000 pycnidiospores were measured.

A pure culture of an isolate of the pathogen was mailed to Commonwealth Agricultural Bureau (CAB, International Mycological Institute (CMI), Surrey, England, for confirmation of the identity of the pathogen. Stock cultures of the fungus on PDAS slants were also stored in the incubator at $15^{o}C$.

4.1.3 Pathogenioty test

Five plates contining 7-day old cultures of the fungus were flooded with sterile distilled water and the conidia harvested by gently scaping the surface of the medium with glass rod, to dislodge the pycnidia. The surface of the

agar medium was then thoroughly rinsed with sterile distilled water. The suspension was blended in a Warring blender for 30 seconds and filtereed through two layers of muslin cloth into a becker.

The inoculum density of the resultant pycnidiospore suspension was determined using the haemocytometer and adjusted to 1 x 10^6 conidia/ml. Using a hand-held sprayer,the pycnidiospore suspension were sprayed to run-off on the leaves of 4-week old kenaf plants raised in plastic pots. The plants were then covered with moistened polyethylene bags and transferred to the glasshouse. Thepolyethylene bags were removed to sprinkle the leaves with water and the bags immediately replaced at 0700 hours in the morning and 1800 hours in the evening for three days after which the polyethylene bags were finally removed. Observations for the symptoms of the disease were carried out daily. Re-isolation of the pathogen on PDAS was done 5 days after the appearance of definite disease symptoms.

4.2 Results and Discussion

4.2.1 Symptomatology

The first symptom of the disease observed, consisted of a greenish, water-soaked spot usually at the leaf margin,

although it could also occur on any part of the leaf surface. The lesion expanded rapidly and later developed a dark brown area at the centre. Examination of the lesion with the aid of a hand-lens (X10) magnification) revealed that the dark brown colour of the central portion was associated with formation of numerous pycnidia.

The dark brown area expanded, just like the water-soaked margin; and the expansion was usually inthe direction of the petiole. The affected leaf withered, but leaf fall only occurred when lesions extended up to the petiole (Plate 3). After heavy rains, as are experiencedin August and September in Mokwa, a severe infection of the upper part of the stem usually resulted in stem die-back (Plate 4). Numerous pycnidia occurred on such stem lesions. The symptoms observed in the present study are similar to that described by Khatua and Maiti (1977), with the exception of the die-back which was not reported by Khatua and Maiti (1977).



Plate 3. Typical lesions of Coniella leaf spot of kenaf.

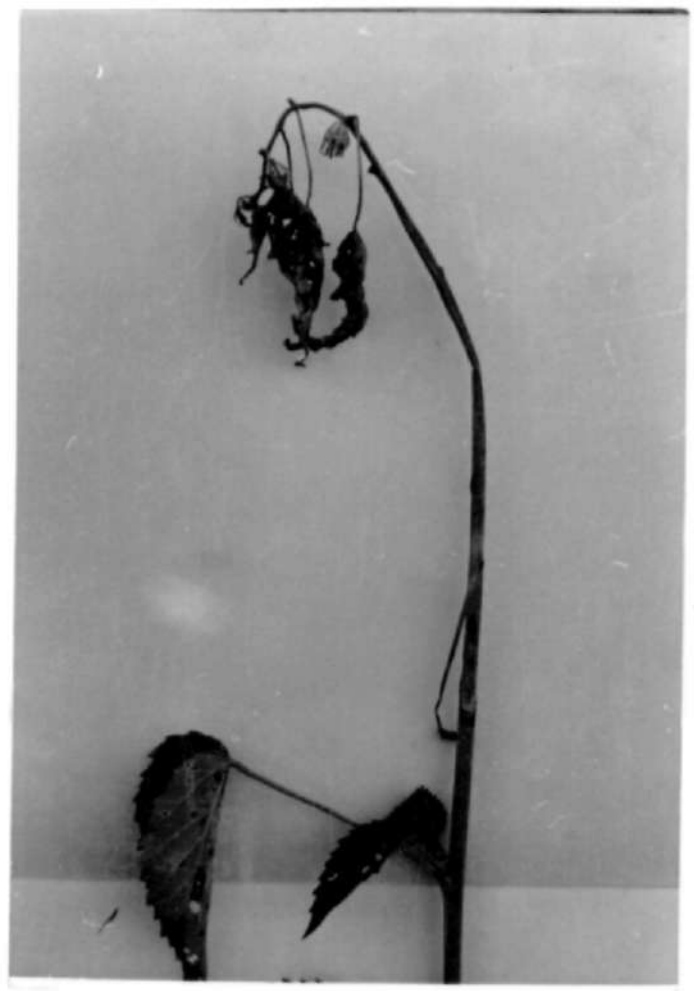


Plate 4. Stem die-back due to severe infection of kenaf stem by C.musaiaensis var hibisci

4.2.2 Isolation and identification of the pathogen

In culture, the organism grew well on PDAS. The colony was initially whitish but it later became dark brown with the production of pyonidia. It completely covered the plate (9.0 cm) in 7 days. The culture grew in concentric rings (Plate 5) when exposed to alternating light and dark periods but they were uniformly brown when grown in continuous darkness (Plate 6).

Microscopic observation showed that the pycnidia were dark brown, globose and ostiolate and measured between 142.5 - 154.2 /um in length and 113.25 - 124.7 /um in diameter. The conidia were hyaline, elliptical, unicellular and slightly elongate. They were rounded at both ends but the middle was somewhat wider; mean size of the conidia was between 3.4 - 7.6 x 10.6 - 14.8 /um. These sizes are in close proximity with those reported by Khatua and Maiti (1977) who first described the organism in India and reported that the pycnidia and thepycnidiospores of their isolate measured between 119.9 to 150.7 /um and 11.75 - 21.15 x 2.35 - 4.65 /um, respectively. A brief description of the organism is stated in appendix V.



Plate 5. Culture of C.musaiaensis var hibisci formed in plates incubated in daily alternative cycles of light and darkness (Agar in the clear area was removed for subculturing before the plate was photographed).

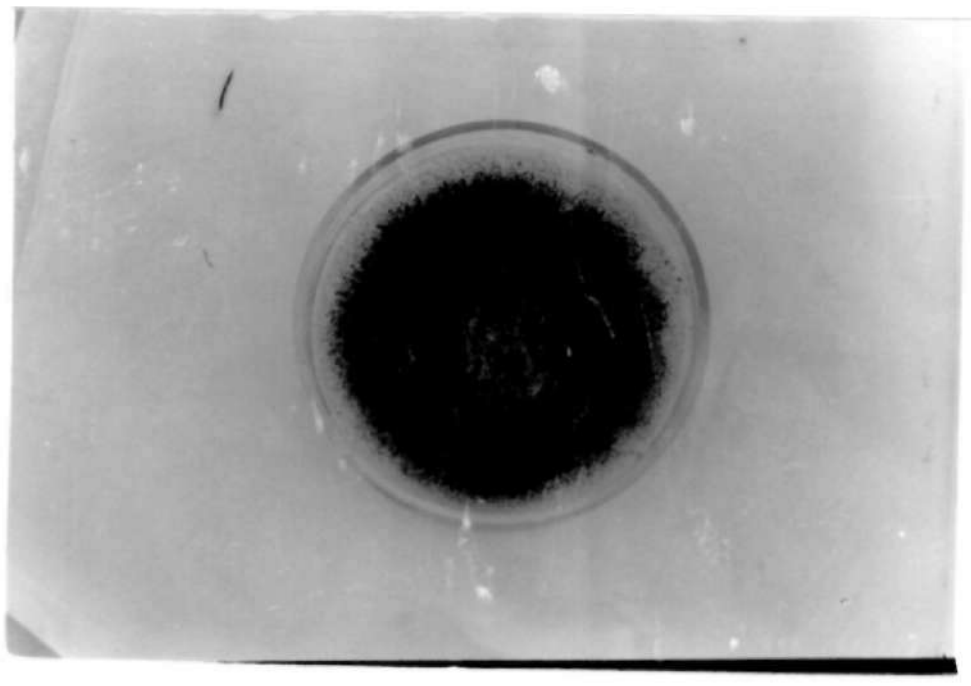


Plate 6. Uniformly brown colony of C.musaiaensis var hibisci produced in plate incubated under continuous darkness.

It appears this is the second record of the organism on kenaf in Nigeria; the first was by Emechebe (1980) who reported Coniella hibisci Morgan-Jones and Coniella musaiaensis B. Sutton on kenaf. The identity of the pathogen was confirmed by CMI, Surrey, London to be Coniella musaiaensis B. Sutton var hibisci B. Sutton (IMI No. 280998 and No.280999b, see Appendix 1).

4.2.3 Pathogenicity test

Pathogenicity test confirmed that C.musaiaensis was the incitant of the disease observed on the kenaf plants. The first symptom of the disease was observed on the artificially inoculated plants as from the fourth day of inoculation as a clear area on the leaf which later became water-soaked. It was also observed that deep brown specks (caused by pycnidial formation) developed at the centre of the water-soaked spots at 7 days after inoculation. Unlike the naturally infected leaves where most of the lesions develop from the leaf margin, the lesions in the artificially inoculated plants developed on any part of the leaf surface.

CHAPTER 5

5. SURVEY OF THE INCIDENCE AND SEVERITY OF CONIELLA LEAF SPOT OF KENAF

5.1 Materials and Methods

5.1.1 Area Surveyed

The survey for Coniella leaf spot of kenaf was carried out in parts of southern Guinea savanna zone of Nigeria in the first week of September, 1984. The places visited were in Niger and Kaduna States, where kenaf is grown extensively for vegetables and other domestic uses. This area lies between 7^{o} and 11^{o}N (Fig. 1).

5.1.2 Survey methods

Since the disease begins to appear in the field as from July, the survey was carried out in the first week of September, 1984 when the disease was very prevalent in the field. Visits were made to kenaf farms in five towns each in Niger and Kaduna States and three farms on the outskirt of each town were selected at random for sampling. The sampled plots in each field were selected randomly. The size of each plot was 5m x 2m, but the number of plots selected in various farms varied. The size of the farm was first estimated; moving to the centre of the farm, five quardrats were thrown randomly to cover the whole farm. At the site of each quardrat, a 2 x 5m plot was marked out and all the kenaf plants within the plot were assessed for disease incidence and severity.

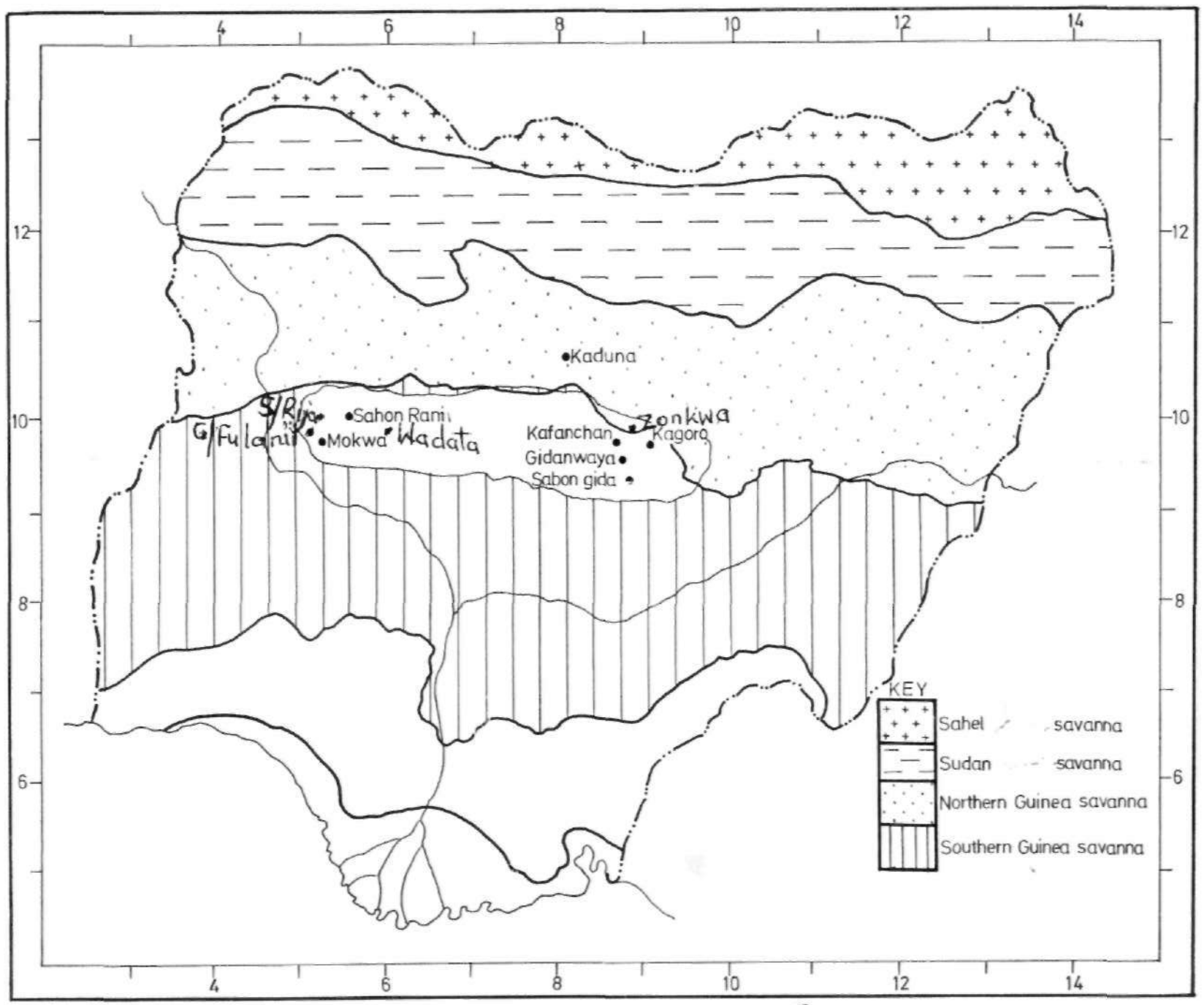


Fig. 1. Map of Nigeria showing the savanna ecological zones and the area surveyed for Coniella leaf spot of Kenaf induced by C. musaiaensis

5.1.3 Disease assessment

The incidence and severity of the disease were visually assessed in all the farms visited. The disease incidence was estimated by counting the number of infected plants in each plot and expressing this as a percentage of the total number of kenaf plants in the plot. The disease severity in each plot was obtained by giving individual plants within the plot numerical disease ratings, as described below, and then calculating the average score for all the plants.

Disease score	Percentage of leaves with leaf spots
1	0
3	1 - 20
5	21 - 40
7	41 - 70
9	more than 70

5.2 Results and Discussion

The survey revealed that most farmers plants Cuba 108 cultivar which had been introduced to them by the Ministry of Agriculture in 1969. The farmers subsequently produced and used their own seeds. It was also observed that the farmers cultivate kenaf mainly for home consumption. The fibre was used for making ropes and for preparing sorghum stalks for fencing. The fibre was also used for tying sticks and grass

TABLE 1: Incidence and severity of Coniella leaf spot of kenaf on selected farm in parts of Niger and Kaduna States in 1984.

Locality	Farms visited in locality	Disease incidence/ site (%)	Disease incidence/ town (%)	Disease Score/ Site	Disease Score/ town
Niger State					
Mokwa	First	95.0 (77.08)	76.09	4.8	
	Second	92.5 (74.11)		7.2	6.27
	Third	95.0 (77.08)		6.8	
Wadata	First	98.0 (81.87)		5.6	
	Second	92.8 (74.44)	77.796	7.8	6.53
	Third	95.0		6.2	
Sabon Rija	First	90.6		7.4	
	Second	92.5 (74.11)	72.146	7.8	7.23
	Third	88.5 (70.18)		6.5	
Gidan Fulani	First	98.0 (81.87)		7.2	
	Second	92.0 (73.57)	75.166	4.6	5.67
	Third	96.4 (70.06)		5.2	
Sabon Rami	First	90.5 (72.05)		5.6	
	Second	98.2 (82.29)	76.183	5.4	5.73
	Third	92.6 (74.21)		6.2	
SED ±			3.494		0.394
LSD (0.05)			3.60		1.43

Table 1 (Cont'd)

Locality	Farms visited in locality	Disease Incidence/ Site (%)	Disease incidence/ town (%)	Disease Score/ site	Disease Score/ town
Kaduna State					
Zonkwa	First	62.5 (55.24)		7.2	
	Second	42.8 (40.86)	44.403	6.1	6.966
	Third	41.5 (40.11)		7.6	
Kagoro	First	68.5 (55.86)		7.3	
	Second	62.5 (52.42)	55.55	7.6	6.766
	Third	72.5 (58.37)		5.4	
Kafanchan	First	92.3 (73.80)		4.6	
	Second	85.6 (67.70)	71.086	4.8	5.4
	Third	90.2 (71.76)		6.8	
Gidan Waya	First	90.5 (72.05)		7.2	
	Second	86.3 (65.28)	59.86	6.5	7.43
	Third	45.2 (42.25)		8.6	
Sabon Gida	First	69.5 (56.48)		7.6	
	Second	82.4 (65.20)	66.476	8.6	7.8
	Third	95.5 (77.75)		7.2	
SED±			8.14		0.907
LSD (0.05)			13.25		0.935

Figures in parenthesis are the angular transformation of the original reading.

CHAPTER 6

6. SCREENING KENAF CULTIVARS FOR RESISTANCE TO CONIELLA LEAF SPOT

6.1 Materials and Methods

6.1.1 Screening under field conditions

The trial was carried out in 1984 and 1986 at Mokwa. Each of the 57 available cultivars in the germplasm collection at Samaru was planted out in the field by middle of May in two rows of 5m each. There was 50 cm space between each row; the spacing between stands on the row being 20 cm. After every four test cultivars, Cuba 108 (a susceptible variety serving as an indicator) was planted. The test cultivars were planted in three replicate being separated from the other by one metre of bare ground.

The test cultivars were artificially inoculated by placing four naturally infected leaves from previous year's crop in the axils of the upper leaves of the last two plants at the edge of each cultivar at four weeks after sowing. At six weeks after sowing, the cultivars were further inoculated artificially by spraying with conidia suspension (1×10^6 conidia/ml) of C.musaiaensis obtained from 7 day-old culture. The inoculation was carried out between 18.00 and 20.00 hours.

The cultivars were assessed for disease severity at 3 weeks after inoculation with conidial suspension. The grading of cultivars for their reaction to attack by Coniella leaf spot was based on resistant (R), high resistance (HR), moderately resistant (MR), moderately susceptible (MS) and susceptible (S), where: 1 = Resistant, i.e. cultivar showed no symptom of disease attack. 3 = High resistance, i.e. cultivar exhibited small, brownish necretic spots, usually with no water soaked margin; the spots are generally few in number affecting fewer leaves (less than 20 percent) in the case of Coniella leaf spot. The MR - cultivars (scored 5) showed typical Coniella leaf spot symptoms (see section 4.2.1) on many leaves (between 21 and 40 percent of total number of leaves) on the plant. The MS - cultivars (scored 7) also showed typical Coniella leaf spot symptoms on between 41 and 70 percent of total number of leaves on the plant while any plant showing typical Coniella leaf spot on more than 70 percent of its total number of leaves was regarded as susceptible and scored 9.

6.1.2 Screening in the glasshouse

The 57 kenaf cultivars in the germplasm collection were also evaluated in 1988 in the glasshouse for their reaction to infection by C. musaiaensis. Plastic pots (19 x 16cm)

were filled with flame-sterilized soil and watered. Seeds of each cultivar were sown in four pots and two weeks after germination, the seedlings were thinned down to two per pot. Cuba 108 cultivar was similarly planted in four pots to serve as a susceptible checked for the trial.

At four weeks after germination (i.e. at 5 weeks after sowing), the above ground parts of the seedlings in each of the four pots of each test line were sprayed to run-off with pycnidiospore suspension (1 x 10^6/ml) of 9-day old cultures of C.musaiaensis grown on PDAS as described above in 4.1.3. The plants were observed for disease development for 10 days. The disease severity was determined on the 10th day of inoculation using the method described above in 5.1.3.

6.2 Results and Discussion

Result of the field screening (Table 2) indicated that the 57 kenaf cultivars exhibited varying reactions to inoculation by the isolate of C.musaiaensis. It was observed that 37 different cultivars were consistent in their reactions


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Table 2. Field and glasshouse reactions of kenaf cultivars to inoculation with Coniella musaiaensis in 1984, 1986 and 1988.

Serial No.	Variety	Reaction in the field in: 1984	1986	Reaction in the glasshouse in 1988
1	Tianung No.1	1.0	1.00	1.0
2	S72-15-2	4.2	4.57	4.5
3	S72-1-17	4.03	3.27	5.5
4	Cuba 2032	1.5	4.86	5.5
5	Cuba	6.13	6.40	6.5
6	S72-78-18-3	6.8	5.50	7.5
7	S72-78-18-10	5.4	3.93	4.5
8	S69-J-13	5.1	5.20	6.5
9	S72-45-9	6.1	4.67	7.5
10	A60-248	5.17	6.43	5.5
11	A60.282	1.0	1.00	1.5
12	A60-511	3.0	3.03	6.0
13	A60-280	4.56	4.83	5.5
14	Everglade 71	4.8	4.30	4.5
15	Everglade 41	3.167	6.37	6.5
16	GR 25/63	3.30	2.03	4.5
17	Guatemala 48	2.03	2.83	6.5
18	BG61/53	3.53	3.30	5.5

Table 2 (Cont'd)

Serial No.	Variety	Reaction in the field in: 1984	1986	Reaction in the glasshouse in 1988
19	BG 58/7	4.96	4.87	**3.5**
20	3789-50	4.20	2.33	**6.5**
21	3431-29	3.43	3.90	**7.5**
22	1892-10	2.63	3.83	**7.0**
23	J69163	2.27	3.70	**6.0**
24	J6987	3.9	2.67	**6.5**
25	Taining 1	1.43	3.30	**3.0**
26	Taining 2	6.13	5.93	**4.5**
27	Balaglade No.7	6.40	7.17	**8.0**
28	Guatamala 45	5.83	5.53	**7.5**
29	25-ASM	6.40	6.37	**7.0**
30	HZ-1	7.20	8.01	**8.0**
31	Hallmalvia	3.83	4.27	**5.5**
32	Local line 1	3.08	2.50	**2.5**
33	Local line 2	2.20	2.70	**4.0**
34	Local line 3	1.63	2.70	**5.5**
35	Local line 4	6.99	6.17	**6.5**
36	Local line 5	5.03	4.29	**7.0**
37	Kenaf	7.3	5.40	**7.5**
38	Kenaf AU-24	7.2	6.27	**8.0**

Table 2 (Cont'd)

Serial No.	Variety	Reaction in the field in: 1984	1986	Reaction in the glasshouse in 1988
39	AU-25	5.07	4.50	8.5
40	Kenaf AU-36	7.0	5.17	7.5
41	Kenaf AU-42	4.63	6.47	7.5
42	Kenaf AU-64	6.4	5.40	7.5
43	Kenaf AU-72	2.3	2.73	6.5
44	Kenaf AU-75	6.27	6.90	8.0
45	Kenaf AU-151	2.07	1.47	5.5
46	Kenaf AU-156	5.67	6.30	7.0
47	Kenaf AU-159	6.40	5.97	8.0
48	Kenaf AU-191	7.9	6.63	8.5
49	Kenaf AU-15	3.37	3.53	5.0
50	AMC 108 Kenaf	1.00	1.00	1.5
51	AC 313	8.23	6.83	8.5
52	Bomo Local	6.70	7.60	7.5
53	HC 583	7.73	7.57	7.0
54	GOD 98-13	7.33	7.53	6.3
55	AC 299	2.33	1.90	5.5
56	Kenaf AU-71	6.33	6.30	8.5
57	Control Cuba 108)	7.30	7.30	7.5
	S.Ed ±	2.20	1.24	2.03
	C.V. %	25.0	25.3	26.2

to field inoculation by the pathogen. Their reaction varied from complete immunity to susceptibility (Table 3). For instance, in the two years (1984 and 1986) three kenaf cultivars (Tianung No.1, A60-282 and AMC-108) appeared to be immune to attack by the Coniella leaf spot. When subjected to direct inoculation by the pycnidiospores of the pathogen in the glasshouse, only Tianung No.1 cultivar was shown to be immune to the pathogen while the other two cultivars showed high resistance (Table 2).

In the two years, seven cultivars - Local lines 2 and 3, Kenaf AM-72, Kenaf AU-151, AC 299, A60-511 and Guatemala 48 consistently exhibited high resistance to inoculation with the pathogen on the field: with the exception of local line 2 which retained this reaction, the other four cultivars were found to be susceptible when subjected to inoculation with the same pathogen under glasshouse conditions.

In the two years also, nine kenaf cultivars exhibited moderate resistance to the disease under field conditions. These are S 72-15-2, S 72-1-17, A60-280, Everglade 71, BG-61/53, BG 58/7, 3431-29, Hallmalvia and Kenaf Au-35. When inoculated in the glasshouse, however, four cultivars, 572-15-2, 572-1-17, Everglade 71 and BG 58/7 retained this

Table 3. Kenaf varieties that exhibited consistent reactions to field inoculation with C.musaiaensis in 1984 and 1986

Immune (I)	High Resistance (HR)	Moderately Susceptible (MS)	Moderately Resistance (MR)	Susceptible
Tianung No.1	Local line 2	S72-15-2	S72-78-3	BZ-1
A60-282	Local line 3	S72-1-17	Cuba	HC 583
AMC-180	Kenaf AU-72	A60-280	S69-J-13	GCD 98-13
	Kenaf AU-151	Everglade 71	A60-248	
	AC 299	BG 61/53	Taining 2	
	A60-511	BG 58/7	Guatemala 45	
	Guatemala 48	3431-29	25-A5M	
	Hallmalvia		Local line 4	
	Kenaf AU-35		Kenaf AU-64	
	Kenaf AU-72			
	Kenaf AU-156			
	Kenaf AU-159			
	Kenaf AU-71			
	S72-78-18-3			
	A60-284			

reaction while cultivar 3431-29 was found to be susceptible and the remaining four - A60-280, BG 61/63, Hallmalvia, Kenaf AU-35 - were found to be moderately susceptible.

In the two years of field screening, 15 kenaf cultivars - 572-78-3, Cuba, "69-J-13, A60-248, Taining 2, Guatemala 45, 25-A5M, Local line 4, Kenaf AU- , AU-12, AU-156, AU-159 and AU-71, A60-284 and 572-78-18-3 - were consistently moderately susceptible under field conditions. Three cultivars - BZ-1, HC583 and GCD 98-13 were also found to be susceptible under field conditions inthe two years.

Erratic reactions were also exhibited by twenty cultivars in the two years (see Table 4). For instance, two cultivars - Evergalde 41 and Kenaf AU-42 - which were moderately resistant under field conditions in 1984 were found to be moderately susceptible in the field in 1986. In contrast, four kenaf cultivars - Local line 5, S72-78-18-10, S72-45-9 and Kenaf AU-25 - which were moderately susceptible in 1984 exhibited moderate resistance in 1986. Five kenaf cultivars - kenaf AU-24, Kenaf AU-36, AU-19, AC 313 and Kenaf - which were susceptible under field inoculation in 1984 were found to be moderately susceptible in 1986. Also two cultivars (Bomo Local and 3 alaglade No.7) found to be moderately susceptible in the field in 1984 (Table 4) were very

Table 4. Kenaf varieties that exhibited inconsistent reactions to field inoculation with *C. musaiaensis* in 1984 and 1986.

Varieties	Reactions 1984	Reactions 1986	Varieties	Reactions 1984	Reactions 1986
Everglade 41	MR	MS	1892-10	HR	MR
Kenaf AU-42	MR	MS	J69163	HR	MR
Local line 5	MS	MR			
S72-78-18-10	"	"			
S72-45-9	"	"			
Kenaf AU-25	"	"			
Kenaf AU-24	"	MS			
Kenaf AU-36	"	"			
Kenaf AU-19	"	"			
AC 313	"	"			
Kenaf	"	"			
Bomo Local	MS	S			
Balaglade No.7	"	"			
GR 25/63	MR	HR			
3789-50	"	"			
Local line 1	"	"			
J6987	"	"			
Cuba 2032	MR	MS			

KEY:

HR = High resistance
MR = Moderately resistant
MS = Moderately susceptible
S = Susceptible

susceptible in the field in 1986. Four kenaf cultivars, GR 25/63, 3789-50, J 6987 and Local line 1, which exhibited moderate resistance under field conditions in 1984 showed high resistance in 1986.

On the other hand, three kenaf cultivars - Cuba 2032, 1892-10, and J 69163 - which showed high resistance in 1984 were moderately resistant in 1986 (Table 4). It was also observed that when the 57 kenaf cultivars were subjected to inoculation with the pycnidiospores of the Coniella musaiaensis in the glasshouse, all but 11 cultivars were either moderately susceptible or very susceptible to attack. Among the eleven cultivars, Tianung No.1 was found to be resistant while cultivars A60-282, Local line 1 and AMC-108 kenaf exhibited high resistance. The remaining seven cultivars, (namely, S72-15-2, S72-78-18-10, S 69-J-13, Everglade 71, GR 23/63, BG 58/7, Taining 2 and Local line 2) were moderately resistant to the pathogen.

When the reactions in the glasshouse were compared with those in the field, it was found that Tianung No. 1 was consistently resistant in the two environments. It was also observed that three cultivars (Local line 1, AMC 108 kenaf and A60-258) consistently exhibited high resistance in the two environments while three cultivars; S72-15-2, Everglade 71 and BG 58/7 showed moderate resistance in both environments.

The detection of higher number of moderately susceptible and susceptible cultivars in 1984 (see Table 2) when compared with 1986 might be associated with higher and better distributed rainfall in 1984 (Appendix III).

While 30 and 26 kenaf cultivars were either moderately susceptible or susceptible to field inoculation in 1984 and 1986 respectively, 45 cultivars exhibited similar reactions (Table 2) in the glasshouse in 1988. This higher number of susceptible cultivars in the glasshouse when compared with the field agreed with the work of Okpala (1981) who screened cowpea lines for their reaction to brown blotch induced by Colletotrichum capsici both in the field and in the glasshouse. He suggested that the difference in reactions at the two environments might be due to expression of resistance by seedlings to brown blotch in the glasshouse being controlled by gones different from those that controlled expression of resistance in the older plants under field conditions.

However, several other factors might be responsible for the difierences observed. For instance, there was high concentration of pycnidiospores used to directly inoculate the leaves. Also inthe glasshouse, there were close to ideal conditions for infection and disease development, especially very high relative humidity were provided by covering inoculated plants with polyethelene bags for 3 days. The temperatures

in the glasshouse when the trial was carried out (25.5 - 27°C) were also much higher than the ambient temperatures in 1984 (21.9 - 20.9) and 1986 (22.7 - 21.5) at Mokwa when the trial was on the field. While there was relatively stable condition in the glasshouse during the relatively short period of development of disease (10 days compared to 35 days in the field) the environmental fluctuations inthe field during the longer period between inoculation and disease assessment might have contributed to the higher number of resistant cultivars on the field. Although Okpala (1981) concluded that the glasshouse provided a better environment for detection of resistance than the field, the field data would probably be more meaningful than those obtained in the glasshouse since kenaf is not grown in controlled environment in Nigeria.

The most dependable and relatively cheap control measure is the use of resistant varieties. One of such cultivars - Tianung No. 1 - has been identified in this study. This, along with three other cultivars, Local line 1, AMC 108 kenaf and A60-282, which consistently showed high resistance in both the field and the glasshouse could be useful as sources of resistance genes to the Coniella pathogen in kenaf breeding.

CHAPTER 7

7. EVALUATION OF THE EFFECTS OF SOME CULTURAL PRACTICES ON THE INCIDENCE AND SEVERITY OF CONIELLA LEAF SPOT ON KENAF

7.1 Effect of Date of Planting on the Development of Coniella Leaf Spot of Kenaf

7.1.1 Introduction

Field observations by the author during the surveys (report in Annon., 1983) showed that 85 percent of kenaf farmers usually plant their crops at the on set of rains in April or May. Fifteen percent of them, however, delayed their planting until later in the season when they would have completed planting other crops such as maize, sorghum and millet. Some farmers even plant kenaf under irrigation as vegetables during the dry season. It was observed that the disease incidence and severity varied considerably between the kenaf plants grown at the on set of rains and those planted at various times in the rainy season. On the other hand, kenaf grown during the dry season was found to be free of the leaf spots. A trial was, therefore, set up to determine the most appropriate time for planting kenaf in order to avoid severe attack by the disease.

7.1.2 Materials and Methods

In 1984, kenaf seeds were sown at seven different times, at fortnightly interval, beginning from 9 May; while in 1986, six sowing dates were studied at the same interval, but beginning as from 23 May. The starting date in each year was determined by the time the rains became established (Appendix 3). The sowing dates constituted the treatments which were replicated three times in a randomized complete block design.

The field was first ploughed, and harrowed before single superphosphate was applied by broadcasting at the rate of 45 kg (P_2O_5) per hectare as basal fertilizer. About five seeds were then sown per hole on the flat and seedling were thinned at 3 weeks after sowing to two seedlings per stand. Each plot measured 3 x 5 m and was surrounded by a 1-m border row.

Spot application of N-P-K (60-45-60) fertilizer was carried out at 4 weeks after sowing. Weeding was first by hand pulling at 3 weeks after sowing, and later by hoeing at 6 weeks when the plants were about 0.5m tall.

For each planting date, the following observations (taken at 50% flowering) were recorded: disease severity, age of the plants at appearance of the first symptoms of the

disease, and plant vigour (height and butt diameter). The number of capsules per plant, number of seeds per capsule, the grain yields were obtained after harvest.

To obtain the fibre yield, plants within 1 m^2 at the centre of each plot were cut at soil level at flowering to ensure maximum fibre yield (Dempsey, 1964a). The stems were then retted for fibre as described in Section 3.3.5 above. Kenaf cultivar Cuba 108 was used for the trial.

7.1.3 Results and Discussion

Results obtained in 1984 (Table 4) and in 1986 (Table 5) consistently showed that the later the planting date, the greater the severity of the disease in the subsequent crop. Thus, June-sown crop showed significantly (P = 0.05) greater disease attack than May-sown crop. In the two years, the disease appeared significantly (P = 0.05) later on kenaf sown in May or June when compared with plantings done as from July. Since kenaf is photoperiod sensitive, flowering when the day length falls below 12½ hours (Dempsey, 1975) the May-and June-sown crop enjoyed longer period of growth before flowering set in in August, hence they were significantly (P = 0.05) taller and produced significantly more grain and fibre yields than the plantings carried out as from July.

Table 4. Effect of date of planting on Coniella leaf spot and yield of kenaf cultivar 108 in Mokwa - 1984

Sowing date	Average disease Score	Plant age at first appearance of symptom (days)	Plant height (m)	Fibre Yield (kg/ha)	No. of capsules/ plant	No. of Seeds/ capsule	Grain yield (kg/ha)
May 9, 1984	2.0	86.67	2.669	2992.99	36.899	18.63	2726.68
May 23, 1984	2.17	82.0	2.497	2732.02	28.47	18.73	2586.68
June 6, 1984	5.03	62.0	2.74	1932.18	30.37	21.67	2353.45
June 20, 1984	7.4	50.0	2.45	1677.38	21.87	20.23	1617.9
July 4, 1984	8.2	33.67	2.295	1296.19	19.10	20.03	1355.56
July 18, 1984	7.7	29.67	1.785	812.60	12.67	16.97	722.23
August 1, 1984	8.23	23.33	1.625	516.28	14.83	19.93	455.56
SED ±	1.003	3.924	0.840	242.98	4.74	1.110	183.54
LSD (5%)	2.180	8.550	1.83	529.46	10.32	ns	347.45

Table 5. Effect of date of planting on Coniella leaf spot and yield of kenaf cultivar Cuba 108 in Mokwa - 1986

Sowing date	Average disease Score	Plant age at first appearance of symptom (days)	Plant height (m)	Fibre yield (kg/ha)	No. of capsules/ plant	No. of Seeds/ capsule	Grain yield (kg/ha)
May 23, 1986	2.4	81.3	2.635	2824.55	39.2	19.3	2957.33
June 6, 1986	5.63	69.3	2.77	2870.7	32.33	21.36	2653.59
June 20, 1986	7.26	55.3	2.42	2170.03	23.73	20.73	1432.34
July 4, 1986	7.13	37.3	2.30	1334.02	14.02	18.57	1299.9
July 18, 1986	7.53	31.3	1.84	816.67	10.97	19.60	848.25
August 1, 1986	7.46	26.6	1.517	483.3	11.07	19.3	491.22
SED ±	0.582	3.514	0.1412	211.969	2.430	0.783	140.266
LSD (5%)	1.297	7.83	0.315	472.27	5.414	ns	312.513

However, there was no significant difference (P = 0.05) among the number of seeds per capsule in both the early and later-planted crops in the two years, despite the difference in disease severity.

7.1.4 Correlation analysis

There was a significantly high negative correlation (r = 0.86 and -0.76 in 1984 and 1986, respectively) between the disease severity and the age of the plant at first appearance of the disease symptoms (Table 6 and 7) in each of the two years. On the other hand, there was a significantly high positive correlation in 1984 (r = 0.69, r = 0.76 and r = 0.86) between the age of the kenaf plant at first appearance of the disease symptoms and the plant height, fibre as well as grain yields. Similarly, significantly high positive correlation (r = 0.84, r = 0.93 and r = 0.94) was observed in 1986 between the age of the plant at disease appearance and the three parameters.

Significantly high positive correlations (r = 0.77, r = 0.68, r = 0.83 and r = 0.88, r = 0.77, r = 0.86 in 1984 and 1986 respectivly) were also recorded between the plant height and number of capsules per plant, the fibre as well as seed yields respectively for each of the two years. While there

Table 6. Correlation between Coniella leaf spot severity score and kenaf yield components at Mokwa in 1984

		I	II	III	IV	V	VI
I	Disease	1.0					
II	Age of plant at first symptom appearance	-.858*	1.0				
III	Plant height	-.601*	.688*	1.0			
IV	Fibre yield	-.797*	.759*	.774*	1.0		
V	No. of capsules/ plant	-.702*	..724*	.683*	.774*	1.0	
VI	Grain yield	-.779*	.864*	.828*	.882*	.421	1.0

* significant at 5% level of probability

Table 7. Correlation between Coniella leaf spot severity score and kenaf yield components at Mokwa in 1986.

		I	II	III	IV	V	VI
I	Disease score	1.0					
II	Age of plant at first symptom appearance	-.757	1.0				
III	Plant height	-.41	.844*	1.0			
IV	Fibre yield	-.615*	.929*	.88*	1.0		
V	No. of capsules/plant	-.776*	.94*	.769*	.387	1.0	
VI	Grain yield	-.743*	.938*	.86*	.343	.382	1.0

* significant at 5% level of probability.

was significantly high positive correlation (r = 0.77 and r = 0.88) between the fibre yield and the number of capsules per plant and seed yield in 1984 the low negative correlation between the fibre yield and the two parameters were non - significant (P = 0.05) in 1986.

The lower disease severity in May-sown crop might be due to low inoculum level of the pathogen in the field early in the season. This observation agreed with that of Tologbonshe (1983) who reported low incidence of Cercospora leaf spot on early planted egg plant, Solanum spp., and attributed this to drier weather and low inoculum level early in the wet season.

The significance of low inoculum level on the field early in the season in disease control was highlighted by Berger (1977) who reported that elimination or reduction in the initial inoculum or delay in its appearance minimises losses due to the disease. The reduction in initial inoculum can be brought about through field sanitation - removal of old plant thrash from the field before planting.

The subsequent increase in disease severity in later-sown crop might be as a result of additive effect of secondary inoculum produced in the earlier-sown crops some of which had already been attacked by the disease at times when the later-sown crops were still at the seedling stage. The increase might also be as a result of the vegetative growth stage of the

later-sown crops coinciding with the overcast weather associated with the heavy rainfall and strong winds at that time of the year (as observed by the author). These conditions probably favoured both increased sperulation and the subseqnet dispersal of the conidia.

The higher grain and fibre yields obtained in the early-sown when compared with later-sown crop was probably because the former escaped early disease attack and grew vigorously. The beneficial effect of early planting in disease control has been reported by Tologbonshe (1983) who obtained higher fruit yield on egg plant, Solanum spp and attributed this to escape of the early-sown crop from attack by Cercospora leaf spot. Jain et al. (1985) also reported that early-sown roselle crops with longer period of vegetative growth maintained higher levels of yields while Oyekan (1977) observed that early season maize escaped severe infections by major diseases, such as Curvularia leaf spot and Helminthosporium blight.

May-sown crop was signifcantly ($P = 0.05$) taller than July-sown crop. Associated with this was the significant increase in the number of capsules per plant in the early-sown crop. These observations might be due to a number of factors; kenaf is photoperiod sensitive (Dempsey, 1975) and consequently flowers inthe Mokwa area as from August. At this time, the May-sown crop had grown taller and should normally be

expected to bear more capsules than the July-sown crops; the latter were shorter and flowered at much younger age; hence the reduction in fibre and number of capsules. However, apart from the effects of photoperiodism and disease, other factors may affect yield which is a complex trait.

7.2 Effect of N, P, and K Fertilizers on Development of Coniella Leaf Spot

7.2.1 Introduction

Preliminary investigation conducted by the author during the survey for the incidence and severity of the leaf spot showed that the common fertilizers used by the farmers in the kenaf growing areas were nitrogen (N), phosphorus (P) and potassium (K). Some farmers applied these nutrients in the following forms: N as urea, P in form of single super phosphate, and K in form of murate of potash. However, very many farmers applied the three minerals in form of 15-15-15 compound fertilizer.

It was also revealed that the type and quantity of fertilizer applied were determined by fertilizer availability and the purchasing power of the farmer; although NPK compound

fertilizer was the most commonly used. It also appeared that the rates of fertilizer applications were approximately the same, regardless of the farmer's location, because the farmers had been taught to use small, empty tomato puree tins (approximately 40 gm) as a standard measure.

Since the farmers have been planting the same cultivar which was distributed to them by the Kaduna State Government since 1969 (P. A. Kognet, 1984, Chief Agricultural Officer, Kaduna state, personal communication) the differences in severities of the disease on crops in different locations might be, among others, attributed to the time of planting, location, the soil nutrient status and the strain of the pathogen. A trial was, therefore, established to determine the influence of the different components of NPK fertilizers and their combinations on the incidence and severity of Coniella leaf spot.

7.2.2 Materials and Methods

The fertilizer used were from different sources. Urea (46% N) was used as the source of N, single super phosphate (18% P_2O_5) was the source of P while murate of potash (60% K_2O) was the source of K.

The trial was carried out in 1984 and in 1986. Before planting in each year, composite soil samples were taken from the experimental field at depths of 0 - 15 cm and 15 - 30 cm. These were air-dried, sieved using a 2 mm sieve and analysed for total N, available P and exchangeable K. The treatments involved a check, sole applications of each of the elements N, P and K and all possible combinations of the elements. There were, therefore, 8 treatments thus; N, P, K, NP, NK, KP, NPK and the check plots to which no fertilizer was added.

Each treatment was replicated four times in a randomised complete block design with each plot measuring 5 by 3 m. A border row of 1 m separated the plots. Kenaf (Cuba 108 cultivar) was planted, 3 seeds per hole, at 20 cm within row and 50 cm inter row. The plants were later thinned to two per stand at 2 weeks after sowing.

The appropriate fertilizer treatment was applied to the plots at 3 weeks after planting by drilling a hole 15 cm from the base of individual stand and carefully working in the fertilizer before covering with soil. Nitrogen was given at the rate of 60 kgN/ha as urea, phosphorus at 45 kgP_2O_5/ha as super phosphate while K was given at 60 kgk_2O/ha as murate of potassium. These are recommended rates for kenaf, Annon, 1983.

7.2.3 Results and Discussion

Plants in plots that received fertilizers (except where only K was applied) grew more vigorously and appeared greener than the check. The study also showed that there were significant differences in the severity of Coniella leaf spot of kenaf as a result of the fertilizer treatments (Tables 8 and 9). The use of combinations of N and P significantly ($P = 0.05$) reduced the severity of the Coniella leaf spot when compared with NPK or N or K in both years. Also combined application of N and P, compared to the check or application of only N, significantly ($P = 0.05$) delayed the number of days to the appearance of first symptoms of the disease on the crop. These two effects of N and P on disease (i.e. lower disease severity and delay in time of appearance of symptoms) were probably one major reason for the greater crop vigour and the significantly higher ($P = 0.05$) fibre and grain yields of the plots that received both elements than plots that received any of the other treatments. The yields might also be attributed to higher efficient utilization of the two elements by the kenaf plant for growth. Such high fibre yields due to combination of N and P have been reported in roselle by Olsen and Dreier (1956) who attributed it to the complementary effect

Table 8. Effect of N, P and K Fertilizer on the development of Coniella leaf spot disease of kenaf in Mokwa - 1984.

Treatment	Disease Score	Plant age at first Symptom appearance	Plant height (m)	Butt diameter (cm)	Fibre yield (kg/ha)	No. of Seeds/capsule	No. of capsule/plant	Grain yield (kg/ha)
NPK (60-45-60)	5.63	55	2.89	4.52	1508.36	19.18	39.65	2617.68
NP	3.3	66	2.73	4.31	1865.85	18.23	45.68	3029.41
NK	3.18	64	2.51	3.71	1533.34	17.28	51.35	2853.44
N	6.35	47.75	2.21	2.30	1108.34	20.05	30.63	2099.48
KP	4.1	59.75	1.64	2.88	937.51	19.3	26.1	1778.99
P	4.02	50	1.55	2.63	957.51	18.7	26.5	1501.93
K	5.15	66	1.17	1.95	948.22	18.8	20.55	1037.89
NO FERTILIZER	3.27	47.75	1.33	1.55	778.26	17.63	19.6	1034.51
SED±	0.570	5.738	0.353	0.380	116.95	1.138	5.973	302.291
LSD (5%)	1.186	11.94	0.73	0.79	243.26	2.367	12.425	143.332

Table 9. Effect of N, P and K Fertilizer on the yield and Coniella leaf spot disease development on kenaf in Mokwa - 1986.

Treatment	Disease Score	Plant age at first Symptom appearance (days)	Plant height (m)	Butt diameter (cm)	Fibre yield (kg/ha)	No. of Seeds/capsule	No. of capsule/plant	Grain yield (kg/ha)
NPK (60-45-60)	6.89	52.25	2.3	4.50	1335.7	45.73	21.4	2315.64
NP	3.1	69.25	2.38	4.88	1285.77	63.92	21.98	3611.54
NK	3.75	69.0	2.21	4.32	1264.36	58.01	18.5	2908.17
N	7.0	40.75	2.00	2.68	1068.21	21.65	19.85	2050.71
KP	4.15	43.5	1.70	3.17	1040.76	17.63	18.13	2064.4
P	4.15	45.0	1.41	3.25	946.16	19.88	20.23	1412.3
K	4.2	58.0	1.80	2.40	1022.77	17.72	18.98	1092.84
NO FERTILIZER	5.40	50.25	1.23	2.18	650.0	10.73	18.60	1129.81
SED±	0.375	4.112	0.227	0.421	84.063	7.509	2.083	244.264
LSD (5%)	0.78	8.55	0.47	0.814	174.85	15.62	ns	508.07

of N and P resulting in increase root proliferation, increased foraging capacity of the plant, and enhanced nutrient absorption.

Except for NP treatments, the combined application of N and K significantly (P = 0.05) increased the number of days to the appearance of the first symptoms of the disease and also significantly increased the grain yields of kenaf when compared with all the other treatments.

The above observations agree with those of earlier workers on roselle (Benson, 1946) and Jute (Das, 1958; Lin, 1959) who reported complementary effects of N and K in increasing yields. The observations also agree with those of Buckman and Brady (1969) who reported that K in the presence of N enhances the development of meristematic tissues. This will in turn result to increase in growth and hence more fibres. Buckman and Brady (1969) also explained that K balances the plant against the excessive useage of N and provides disease resistance.

The use of N as the sole fertilizer or in combination with both P and K appeared to pre-dispose the kenaf plant to early and severe attack by the Coniella leaf spot. However, such plants grew significantly taller than the check and also produced significantly (P = 0.05) higher fibre and grain yields when compared with the check. This observations are in consonance with the report on roselle by Jain, et al (1965)

who reported that high levels of N increased yield and also the succulence of vegetation, giving the leaves such large, thin-walled colls that they were readily attacked by fungal pathogens.

When applied as the sole fertilizer, P did not significantly reduce the disease severity in 1984, nor did it significantly delay the time of appearance of the first symptoms of the disease on the crop in both 1984 and 1986, compared to the check treatment. It appears the application of P alone had little effect on Coniella leaf spot attack on Kenaf. The non-reduction in disease severity due to application of P has been recorded by Lin (1959) and Jain, et al (1965) on jute and roselle, respectively, who suggested that phosphate application seemed to have little to do with disease resistance.

Inconsistent results were obtained with application of K as the sole fertilizer. Thus, it reduced disease severity significantly in 1986 but not in 1984 when compared with the check. By contrast, in 1984, but not in 1986, K application significantly ($P = 0.05$) increased the age of the plant at the appearance of the first symptoms of the disease. The role of K in disease resistance in the present study is thus not clear. This results did not agree with earlier work on stem rot of jute (induced by Rhizoctonia spp.)

by Gosh and Basak (1964) who reported that K increased disease resistance and checked disease incidence. There were no significant differences in grain yields of the kenaf supplied with K in the two years.

The combined application of N, P and K as fertilizer resulted into more severe disease attack in both years on kenaf, compared to the check. There was also early appearance of the disease symptoms where these three elements were combined. It was also observed that the balanced fertilization appeared to increase rapid leaf production and severe disease attack at the same time. The severe leaf spot attack usually resulted in premature leaf fall which was apparently compensated for by increased leaf production towards the plant apex. This apical meristematic activities resulted to taller plants and hence significantly ($P = 0.05$) higher fibre yields when compared with the other treatments (except combined applications of N and P or N and K.).

CHAPTER 8

8. CHEMICAL CONTROL OF CONIELLA LEAF SPOT OF KENAF

8.1 Introduction

The use of fungicides is a quick and effective means of controlling fungal diseases in plants. Since there appeared to be no previous record of work on fungicidal control of Coniella leaf spot of kenaf, a trial was established to evaluate the efficacies of four fungicides for the control of the disease.

The fungicides tested were Delsene M (carbendazim plus maneb), Benlate (benomyl), Dithane M45 (mancozeb), and Calixin M (a mixture of maneb and tredimorph). Their choice was based on the fact that they were readily available in the market as well as with local farmers who cultivate tomato and cowpea in addition to kenaf. The trade and common names of the fungicides as well as their chemical compositions are given in Table 10.

8.2 Materials and Methods

8.2.1 In-vitro assessment of effects of dosage of 4 fungicides on growth and conidia production in C. musaiaensis

The recommended spray concentrations of Delsene M, Dithane M-45, Benlate and Calixin M are 1.2, 2.5, 0.6 and 3.5 kg of commercial products per hectare (ha) respectively. To assess the

Table 10. Chemical nature, trade and common names of fungicides evaluated in the laboratory and field for their efficacies against C. musaiaensis.

Trade name	Common name of fungicidal active ingredient	Chemical nature of the active ingredient
Delsene M	Carbendazim + maneb.	carbendazim = (methyl-zimidazole 2-yl carbamate) 10% maneb (manganese ethylene bisdithio carbamate) - 64%
Dithane M-45	Mancozeb	2.5% Zn ion + 20% Mn es in maneb.
Benlate	Benomyl	methyl 1-(butylcarbamoyl) benzimidazole-2-yl carbamate.
Calixin M	Calixin M	tredimorph (11%) maneb (36%)

fungicides, different proportions of the above dosages were incorporated into 250 ml of potato dextrose agar (PDA). The tested rates for each fungicide were designated as 2x, x, 0.5x, 0.25x and 0.125x per 250 ml of PDA; where x is the above recommended dose of the fungicide to be dissolved in 250 ml of water assuming that the rate per ha is to be dissolved in 200 litres of water.

To prepare PDA-fungicide mixture, the following method was adopted. The approximate weights of the fungicide were added separately into 500 ml conical flask containing 250 ml of molten PDA. The flasks were shaken carefully to ensure even distribution of the fungicides.

To ensure that equal quantity of the medium was poured into each plate, the following methods was adopted. Twenty McCartney bottles were sterilized along with the media in the autoclave. Using one McCartney bottle for each concentration of a fungicide, 24 ml of the concentration (filling the bottle up to a marked point on the neck) was poured into each petri dish. The whole process was carried out in a sterile chamber. Five petri dishes were poured for each concentration. The plates were allowed to set for 24 hours.

One 0.6-cm (diameter) cork borer was used to transfer the fungus from 7-day old culture on to the centre of each plate. Check treatments consisting of PDA plates alone were

similarly inoculated for each fungicide concentration in the experiment. The plates for different concentrations of the fungicides were arranged in completely randomized design on the laboratory bench (temperatures: 24 - 30°C) Fungitoxicity was assessed by measurement of linear (diameter) growth of the fungus after 7 days.

8.2.2 Effect of fortnightly sprays of 4 fungicides on Coniella Leaf spot of Kenaf

The trial was carried out in the wet season (June to September) of 1983. The four fungicides (Delsene M, Benlate, Dithane M-45 and Calixin M) were screened inthe field at Mokwa at the rates of 1.2, 0.6, 2.50 and 3.5 kg per ha respectively, using cultivar Cuba 108. The experimental layout was a randomized complete block design with 3 replicates, each plot measuring 5 m x 5 m. A 1-m border separated plots from each other.

Spraying, started at 4 weeks after sowing, was applied at 14-day interval until the crop flowered (i.e. 11 weeks after sowing). Observations were taken on disease severity, the age of plant at the appearance of the first symptoms of the disease, plant height, butt diameter, grain and fibre yields.

8.2.3 Effect of spraying intervals of four fungicides on Coniella leaf spot and yield of kenaf

The four fungicides (Delsene M, Benlate, Dithane M-45 and Calixin M) were tested in the field at Mokwa in the growing season of 1984 and 1986, starting from June with the aim of determining the most respective spraying intervals for the control of the leaf spot. The experimental layout was a split plot design in which the main plots were the four fungicide treatments and the sub-plots were four spraying intervals of 7, 14, 21 and 28 days. The check consisted of plots sprayed with water at each of the above intervals. In each year, the trial was replicated three times while the sub-plots measured 5 x 5 m. A 1-m border separated the sub-plots, while a 2-m border separated the main plots. Cuba 108 kenaf cultivar was used as test crop.

Spraying was started from four weeks after sowing, and it continued until the first sign of flower initiation was observed between 8 and 15 August. On the whole, the spraying lasted for 10 weeks. Observations were taken onthe disease severity and yield of the crop.

8.3 Results and Discussion

8.3.1 In-vitro assessment of four fungicides for their efficacies against Coniella musaiaensis

The result obtained (Table 11) showed that Delsene M (carbendazim + Maneb) and Benlate (benomyl) completely inhibited mycelial growth of C.musaiaensis in agar plates at their recommended field spray doses (i.e. 3 gm and 1.5 gm of commercial product per litre of water, respectively) (plate 7). Carbendazim was effective in inhibiting mycelial growth at 12.5% of the field rate (i.e. 38 gm of Delsene M per litre). At a corresponding concentration of benomyl (i.e. 19 gm of Benlate 50 WP/litre) benomyl was also effective since it permitted only 16.7% of the growth inthe check (plate 8).

Mancozeb (Dithane M-45) and Calixin M, on the other hand, gave 100% growth inhibition at twice their field spray doses and about 90% growth inhibition at the field spray doses, compared with the growth of the check. Even at 12.5% of the field spray doses, Dithane M-45 and Calixin M still effected mycelial growth inhibition up to 82 and 89% (Plates 9 and 10).

Thus, the most effective fungicides in-vitro were carbendazim and benomyl. These two fungicides incidentally have the highest concentration of active ingredients per kg of commercial products (Benlate, 50% WP of benomyl; Delsene M,

Table 11. Effect of concentrations of Delsene M, Dithane M-45, Calixin M and Benlate on the growth of C.musaiaensis in agar medium

Fungicide	Concentration of fungicides in agar (gm/litre)	Growth of C. musaiaensis 7 days after inoculation	
		Colony diameter (cm)	Growth as % of growth of control
Delsene M	6.00	0.0	0.0
	3.00	0.0	0.0
	1.50	0.0	0.0
	0.85	0.0	0.0
	0.38	0.0	0.0
Dithane M-45	12.50	0.0	0.0
	6.25	0.8	8.89
	3.125	1.06	11.78
	1.563	0.98	10.89
	0.782	1.5	16.67
Calixin M	15.00	0.0	0.0
	7.50	0.86	9.56
	3.75	0.94	10.44
	1.875	1.1	12.22
	0.938	1.	11.1
Benlate	3.00	0.0	0.0
	1.50	0.0	0.0
	0.75	0.0	0.0
	0.38	0.0	0.0
	0.19	1.5	16.67
Control	No Fungicide	9.0	100
	" "	9.0	100
	" "	9.0	100
	" "	9.0	100
	" "	9.0	100

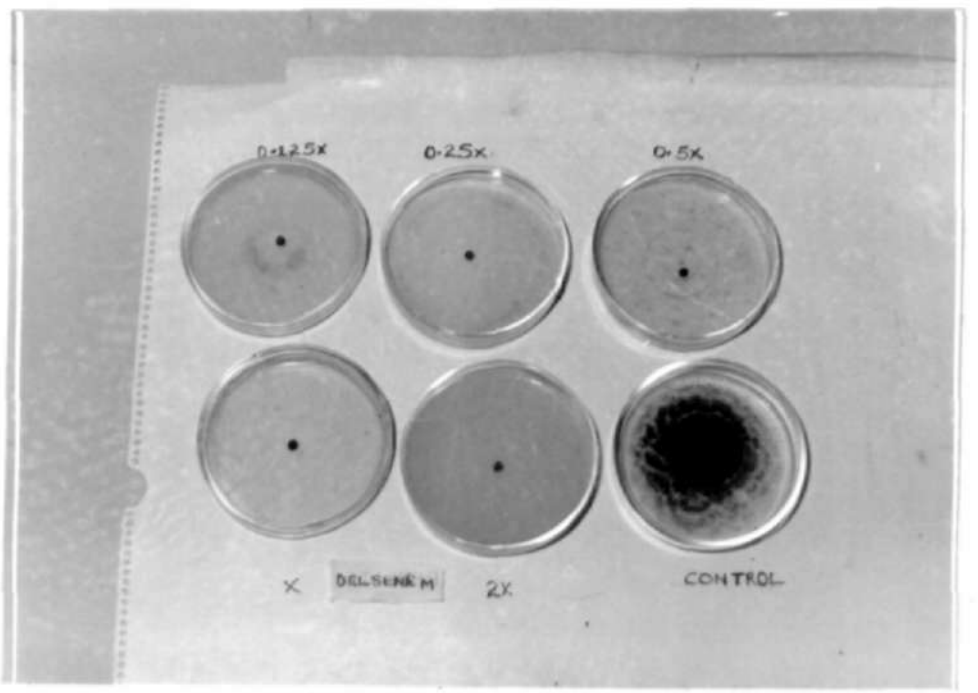


Plate 7. Effect of concentration of carbendazim on the mycelial growth of C.musaiaensis in potato dextrose agar (PDA)

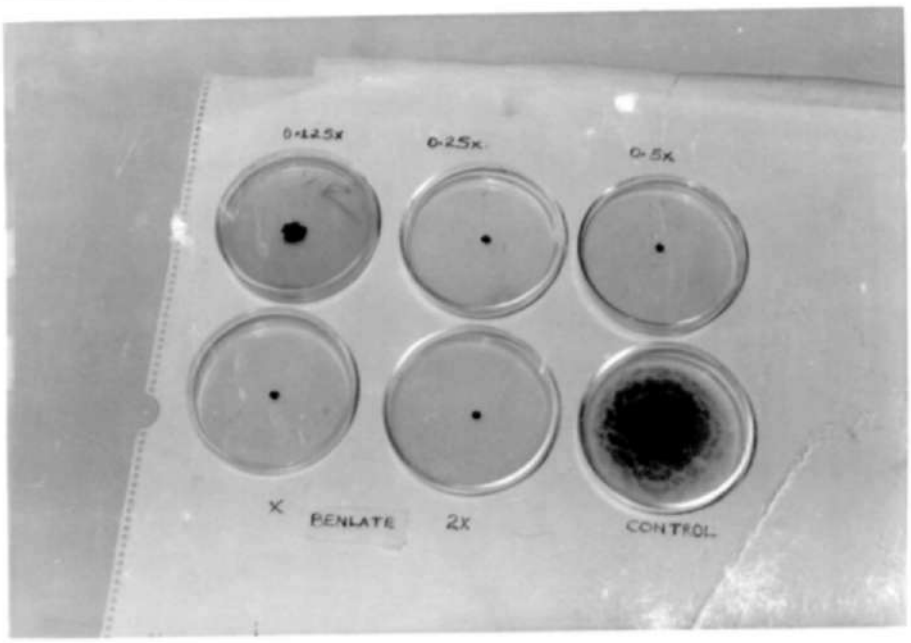


Plate 8. Effect of concentration of benomyl on mycelial growth of C. musaiaensis in PDA medium.

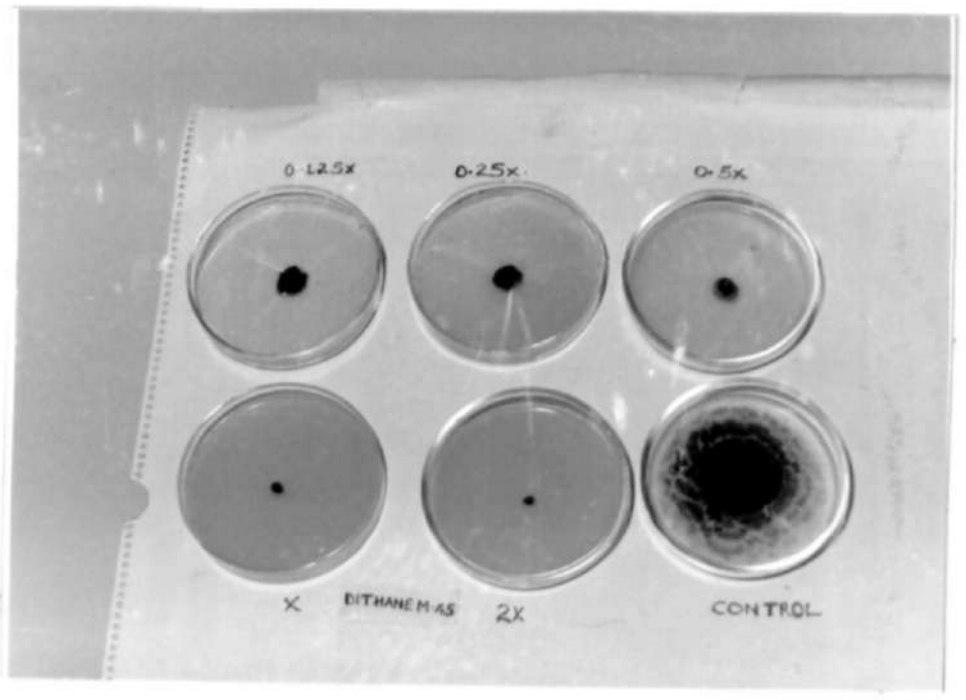


Plate 9. Effect of concentration of mancozeb on the mycelial growth of C.musaiaensis in PDA medium.

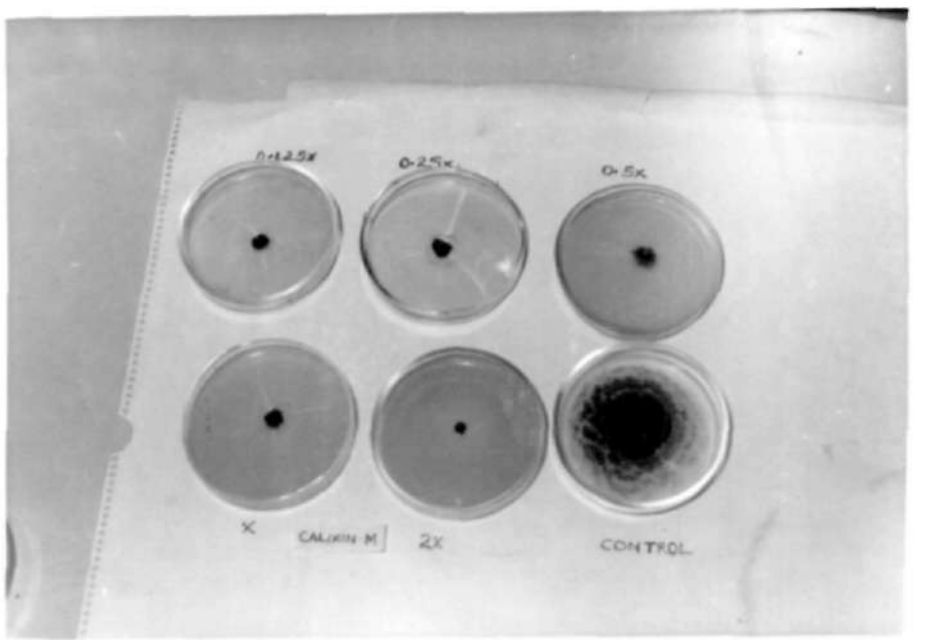


Plate 10. Effect of concentration of Calixin M on the mycelial growth of C. musaiaensis in PDA medium.

10% and 64% carbendazim and maneb respectively). The other two fungicides, Dithane M-45 (22.5% WP mancozeb) and Calixin M (10% and 36% WP tredimorph and maneb respectively) were not effective at the field dosage equivalents, but completely inhibited the mycelial growth at double dosage.

Dithane M-45,Delsene M and Benlate have been shown by other workers to inhibit the mycelial growth of fungi at different concentrations in-vitro (Alafiatayo, 1982 on Colletotrichum capsici; Tologbonshe, 1983 on Cercospora spp). The effectiveness may be attributed to the direct close contact between the fungus and the fungicides in the agar plate. However, the ineffectiveness of a fungicide in inhibiting mycelial growth in-vitro may not necessarily be the same on the field. For instance, it has been demonstrated in classic experiments by Davis and Dimond (1952) that some systemics which are fungicidally inactive in-vitro could initiate antifungal effect in vivo.

8.3.2 Effect of fortnightly sprays of four fungicides on *Coniella leaf spot of kenaf*

Each of the four fungicides (namely, carbendazim, benomyl, mancozeb and calixin M) significantly (P = 0.05) reduced the severity of the Coniella leaf spot in 1983 when sprayed at 14-day interval, compared with the unsprayed check (Table 12). In addition, spraying significantly (P = 0.05) increased the

Table 12. Screening four fungicides for their efficacies in controlling the leaf spot of kenaf in Mokwa in 1983.

Treatment	Disease Severity	Age at appearance of first symptom of disease (days)	Plant Height (cm)	Butt diameter (cm)	Capsule per plant	Seed per capsule	Seed yield (kg/ha)
Delsene M	2.44	73.6	250.66	3.2	33.4	19.579	2356.0
Benlate	3.72	76.8	231.139	2.977	34.06	21.74	2104.0
Calixin M	3.08	65.8	291.9	3.025	30.78	20.9	1572.8
Dithane M-45	3.12	72.4	262.44	2.965	32.7	21.939	1424.0
Control	6.64	41.8	189.079	2.345	24.5	18.86	1127.2
SED±	0.512	2.134	9.537	0.18	2.089	1.4097	260.664
LSD (5%)	1.079	4.451	19.895	0.3916	2.358	2.941	543.75

number of days to the appearance of first symptoms of the leaf spot, when compared with unsprayed check. Both the grain and fibre yields of the sprayed plotswere significantly (P = 0.05) higher than those of the unsprayed check.

Although fortnightly sprays appeared to be a good control of the disease, the number of sprays (5 sprays/season) implies the use of large quantitites of fungicides, the prices of which increase every year. Edgington et al (1980) has noted that systemic fungicide sprays have curative, post-infection action. Hence there is less need to constantly protect the foliage. Also less spraying will reduce the stress on the environment. The fortnightly spraying consumed labour and hence the cost of production became high. The second field trial, therefore, was set up to determine the most appropriate spraying intervals that would still give effective control of the disease inthe field, while at the same time minimizing costs.

8.3.3 Effect of spraying intervals of four fungicides on Coniella leaf spot and yield of kenaf at Mokwa in 1984 and 1986

The results obtained in 1984 (Table 13) and 1986 (Table 14) indicated that weekly sprays of each of the four fungicides were more effective than spraying at other intervals, especially

Table 13. Effect of spraying intervals of four fungicides on the severity of Coniella leaf spot and yield components of kenaf in Mokwa, 1984.

	Spraying interval (days)	Disease Score	Age at appearance of first symptom of disease (days)	Final Plant Height (m)	Butt diameter (cm)	Fibre Yield (kg/ha)	No. of capsules per plant	No. of Seeds per capsule	Grain yield (kg/ha)
Delsene M	7	1.77	76.0	2.69	3.02	4070	38.20	19.77	1850
	14	2.0	65.3	3.14	3.44	3957	39.03	20.97	1781
	21	4.53	56.3	2.85	3.24	3598	33.47	20.33	1304
	28	5.87	50.7	3.06	2.95	3845	27.60	19.70	938
Water spray		6.07	47.0	3.07	2,97	3989	25.00	19.50	871
Benlate	7	1.77	73.0	3.04	2.99	4295	40.73	21.50	1800
	14	2.17	71.7	3.06	2.89	4230	39.93	20.43	1904
	21	4.60	60.0	2.83	2.81	3881	32.80	17.60	1339
	28	6.40	54.7	2.85	3.19	2975	25.87	20.13	1071
Water spray		7.70	47.0	2.29	2.71	3185	24.47	18.32	909
Calixin M	7	3.07	65.7	2.95	2.97	3200.0	39.80	21.53	1951
	14	4.37	55.0	3.05	2.24	3100	37.53	19.80	189.5
	21	6.47	18.0	2.87	2.39	3317	32.53	19.92	150.5
	28	7.47	46.3	2.87	2.32	2677	27.37	18.53	1142
Water spray		7.35	45.7	2.73	2.24	2557	25.83	21.37	1132

Table 13 (cont'd)

	Spraying interval (days)	Disease Score	Age at appearance of first symptom of disease (days)	Final Plant Height (m)	Butt diameter (cm)	Fibre Yield (kg/ha)	No. of capsules per plant	No. of Seeds per capsule	Grain yield (kg/ha)
Dithane M.45	7	2.40	68.7	2.99	3.04	3467	44.30	21.60	1726
	14	3.40	58.3	2.95	3.02	3196	41.43	19.63	1674
	21	4.93	57.7	3.02	3.02	3967	36.03	20.40	1451
	28	6.47	58.3	2.89	2.79	2800	33.70	21.00	1186
	Water spray	6.50	48.6	2.94	2.85	2300	28.77	19.50	1217
SED±		0.627	5.212	0.204	0.2868	240.9	2.917	1.885	129.0
LSD (P = 0.05)		1.831	15.364	0.559	0.929	719.230	8.952	6.613	348.340

Table 14. Effect of spraying intervals of four fungicides onthe severity of Coniella leaf spot of kenaf in Mokwa, 1986

Fungicide	Spraying interval (days)	Disease Score	Age at appearance of first symptom of disease (days)	Final Plant Height (m)	Butt diameter (cm)	Fibre yield (kg/ha)	No. of capsules per plant	No. of Seeds per capsule	Grain yield (kg/ha)
Delsene M	7	1.87	78.0	2.70	3.39	4267	40.37	19.87	1842.0
	14	2.47	76.7	3.12	2.95	3400	33.33	19.47	1735
	21	5.07	63.3	3.08	2.97	3367	30.40	19.83	1276
	28	6.80	45.7	3.03	2.91	3900	30.50	20.80	987
Water spray		7.47	48.0	3.11	2.88	4233	24.20	19.53	1221
Benlate	7	2.20	74.3	2.99	2.91	4500	35.23	19.30	1523
	14	3.67	75.0	3.21	2.92	4167	35.87	19.67	1540
	21	5.87	59.0	2.95	2.89	3533	31.87	18.77	1253
	28	6.80	59.3	2.91	2.72	3100	21.93	19.50	1134.0
Water spray		7.93	51.6	2.39	2.86	4269	21.57	18.90	1094.0
Calixin M	7	3.60	73.7	3.02	2.24	3357	35.83	21.33	1888
	14	3.20	67.7	3.06	2.05	3300	30.40	20.10	1722
	21	5.95	48.7	2.96	3.01	3000	29.43	20.21	1575
	28	8.20	48.7	2.93	2.40	2967	29.07	21.23	1421
Water spray		7.93	51.0	2.96	2.29	2800	28.21	22.97	1351
Dithane M-45	7	2.40	68.7	2.75	3.22	4567	43.67	21.00	1774
	14	3.47	69.7	2.88	2.91	3500	43.58	20.33	1544
	21	5.20	57.7	3.03	3.06	3700	33.40	20.17	1329
	28	7.20	53.0	3.04	2.99	3300	34.63	19.80	1335
Water spray		7.40	46.3	3.05	2.99	3335	29.40	19.37	1284
SED ±		0.5335	5.241	0.1837	0.2391	345.0	2.959	1.454	138.1
LSD (0.05)		1.521	15.186	0.893	0.69	937.97	8.78	3.893	416.74

spraying at 21- or 28-day intervals. Of all the fungicidal spray regimes Delsene M at fortnightly and weekly intervals and Benlate at 7-day interval were the most effective during the two years.

Delsene M contains both systemic (carbendazim) and protectant (maneb) fungicides while Benlate contains only a systemic fungicide. Delp (1980) has recommended the use of companion fungicides with different modes of action as the most effective method of controlling disease so as to avoid the emergence of resistant strains of a fungus. Peterson and Edgington (1971) reported that carbendazim fungicides are readily transported acropetally to the upper parts of the plant where they remain active. The present result agreed with earlier report by Delp (1980) who reported that the use of carbendazim plus maneb has been very effective in controlling Cercospora on sugar beats in Greece.

While benomyl was very effective in controlling the Coniella leaf spot when applied as foliar spray in 1984, it was less effective in 1986. Many factors could be responsible for this. It is possible that the particular pack has been manufactured over a long time, thus, the shelf life might have contributed to its less effectiveness. There are the differences in weather conditions (temperature, quantity and distribution of rainfall) in the two years.

Another reasons might have been the presence of resistant strains of the fungus. Decker (1976) reported that resistance to benomyl application has occurred in many instances either spontaneously on the field or after mutagenic treatments in the laboratory. Delp (1980) reported that the first recorded case of fungicide resistance was to benomyl.

Systemic fungicides, such as carbendazim and benomyl, however, have some advantages. Edgington et al,(1980) reported that they eradicate the pathogens shortly after infection and also get redistributed within the plant and thus compensate for poor coverage. Their disadvantages Edgington **et** al, (1980) include, their relative insolubility when applied as wettable powders which makes it difficult to transverse the leaf cuticle. He estimated that only 5% of a systemic applied to leaves enter while the rest remain as protectants on the leaf surface.

The relative ineffectiveness of manoozeb and Calixin M in controlling the disease in the field agreed with the results of the agar plate tests. Calixin M and mancozeb (with 36 and 20 percent maneb active ingredient respectively) are non - systemic. When applied as foliar sprays, they are expected to be active only at the point of application (Edgington et al, 1980) and hence prevent disease attack in these parts. Thus their

effectiveness at weekly and fortnightly application was due to frequency of application. When applied at 3-week or 4-week interval, intervening prevailing conditions on the field might have affected their efficacies. The sprayings were applied during the raining season and on some occasions, the rain usually fell within a short period of their application. This prevents the adherence of the chemical to the plant as a result of washing off. Woodcock (1971) has reported that increased rainfall prevents uptake and translocation of systemic fungicides and also reduced the effectiveness of non - systemics by gradual attrition of the fungicide deposits by wind and rain water through hydrolysis.

There were significant differences (P = 0.05) among treatments with respect to ages of kenaf plants at time of appearance of the first symptoms of the disease. Thus, benomyl and carbendazim when sprayed at weekly, fortnightly, significantly increased the age of the plants at time of appearance of the first symptoms of the disease in both 1984 and 1986, when compared with the check plots. Applications of mancozeb and Calixin M gave similar results only when they were applied weekly in 1984 and fortnightly in 1986.

The effective spray regimes also gave significantly higher grain yields. Thus plots sprayed with Benlate or

Delsene M at 7- to 14-day intervals or with Calixin M or Dithane M-45 at weekly interval produced significantly higher grain yields than the check plots. These observations implied tht Coniella leaf spot attack has direct effect on the grain yields of kenaf.

In 1984 and 1986, there were no significant differences (P = 0.05) in fibre yields among the plots sprayed with Delsene M at the four intervals and those that were not sprayed with any fungicide. However, in 1984, the fibre yields of plots sprayed with benomyl at weekly or fortnightly intervals were significantly (P = 0.05) higher than those of the check plots while in 1986, there was no significant difference (P = 0.05) in fibre yield between the check plots and the plots sprayed with Benlate, regardless of spraying interval. There was also no significant difference in fibre yields (in the two years) between the plots sprayed with Calixin M, irrespective of spraying interval, and the check plot.

In many cases, the unsprayed plots out yielded those sprayed with fungicides. It appeared the control of Coniella leaf spot by application of fungicides has little or no effect on the fibre yield of kenaf. The implication of this is that if the ultimate aim of planting kenaf is for fibre production, there may be no need to apply fungicides for the control of Coniella leaf spot.

CHAPTER 9

9. DETERMINATION OF ALTERNATIVE HOSTS OF CONIELLA MUSAIAENSIS

9.1 Introduction

Determination of alternative hosts of C.musaiaensis, the pathogen of Coniella leaf spot of kenaf, will be a useful information in understanding its survival during the dry season when the kenaf will not be on the field. Such information will help in the formulation of control measures for the disease that would include adoption of appropriate crop rotation or field sanitation.

Khatua and Maiti (1977), who provided the first report of C.musaiaensis as a pathogen of kenaf also identified roselle, Hibiscus sabdariffa L., as an alternative host of the fungus. Since there appears to be no other reference in the literature about further work on the subject, an experiment was done to study the host range of the pathogen among the crops and other flora commonly found within the kenaf growing areas of Nigeria.

9.2 Materials and Methods

Seventeen species of flowering plants (see Table 15) in nine taxonomic families were screened in the glasshouse for their susceptibility to Isolate-2 (Mokwa isolate) of C.musaiaensis. Three of the plant species (sweet potato,

Ipemea batatas L., cassava - Manihot esculenta Craz and grape vine - Vitis vinifera L.) were raised from stem/vine cuttings. Self -sown Sida sp. seedlings were dug up with as much of the roots and soil as possible and transplanted into pots while the other fifteen species were raised from seeds in heat sterilized soil in 12 cm (diameter) plastic pots in the glasshouse. Six pots (2 seedlings/pot) of each plant were grown.

The plants were inoculated with a suspension of pycnidospora (containing 1 x 10^6 spores/ml) of the isolate four weeks after planting. In each case, two uninoculated seedlings of each species served as the check. The experiment was carried out between August and September of 1986. The severity score was based on disease evaluation scale described in section 5.1.3.

9.3 Results and Discussion

Among the 17 plant species inoculated artificially in the glasshouse, four were susceptible to the pathogen. These belong to two families - Malvacese (cotton, Gossypium hirsitum L.; okra, Abelmoschus esculentus L.; roselle-Hibiscus sabdariffa L.) and Solanacceae (pepper, Capsicum annum L.).

There was a marked variation in susceptibility between the three malvaccus species, with roselle being the most susceptible (Table 15). While the disease developed on the

Table 15. Pathogenicity of C.musaiaensis in some species of crops and weeds in the glasshouse at Samaru

Common Plant name	Scientific name of plant	Family	Disease Severity Score
Spinach	Amarantus viridis L.	Chenepodiaoeae	0.0
Sweet Potato	Ipomoea batatas L.	Convelvulaceae	0.0
Cassava	Manihot esculenta Cranz	Euphorbiaceae	0.0
Sorghum	Sorghum bicolor L (Moench) (cv.SK 5912)	Granineae	0.0
Millet	Pennisetum typhoides L.	Gramineae	0.0
Maize	Zea Mays L. (cv.TEB)	Gramineae	0.0
Cotton	G. hirsitum L.(cv.Samaru 77)	Malvaceae	4.5
Okra	Abelmoschus esculentus L.	Malvaceae	0.0
Roselle (red calyx)	H. sabdariffa L. (var. I)	Malvaceae	5.0
Roselle (green calyx)	H. sabdariffa L. (var II)	Malvaceae	7.0
Sida sp	Sida alba L.	Malvaceae	7.2
Tossa jute	Corchorus capsularis L.	Tiliaceae	0.0
Jute	Corchorus oliontorus L.	Tiliaceae	0.0
Urena	Urena lobata L.	Malvaceae	0.0
Grape vine	Vitis vinifora L.	Vitiaceae	0.0
Pepper	Capsicum annum L.	Solanaceae	0.0
Egg plant	Solanum macrocarpon L.	Solanaceae	3.5
Cowpea	Vigna unguiculata L.(var.Sampea 7)	Papilionaceae	0.0

entire leaf surface, including the margin, in roselle, as it typically does on kenaf, the lesions in cotton and okra did not occur on the margin, although they were in other respects, typical of the foliar lesion in kenaf, In the case of pepper, the lesions were quite different from those of the Malvaceous plants. They appeared as rather small brownish, necrotic spots on the leaf surface and were probably a type of high resistance of the host to the pathogen.

The most susceptible hosts were the two cultivars of roselle. One of the roselle cultivars (with green calyx) is commonly gorwn in the rain forest zone and the middle belt of Nigeria for its fleshy calyx which is cooked and eaten as vegetable. The other variety(with red calyx) is usually grown in the fadamas during the dry season in the northern Guinea savanna for its leaves which are eaten as vegetable.

Khatua and Maiti (1977) who first reported this disease on kenaf also identified roselle as an alternative host of the pathogen. Cotton and okra belong to the Malvaveae family just as roselle and kenaf/and are being reported for the first time as alternative hosts of the pathogen. Out of the two members of Solanaceae family tested, pepper was found to be weakly susceptible. This mild attack on sweet pepper might probably mean that the pathogen has a much wider host range in Nigeria than is presently recognised.

The significance of these alternative hosts is that pepper and cotton can withstand the adverse weather conditions during the dry season, thereby surviving from one season to the other in the field; they could, therefore, serve as potential sources of primary inoculum at the beginning of a new cropping season.

It is suggested that planting of any of the above hosts within or near the kenaf plantation should be avoided. Non-host plants should probably be used as intercrop or as barrier crops around the kenaf field. Berger (1977) has reported that the elimination of the alternative hosts has been effective in controlling rusts induced by Puccinia species. He also reported that planting barrier crops and inter-cropping with non-host plants reduce the rate of arrival of the conidia on the crop and hence slowed down the rate of infection.

Crop rotation by planting non-host crop, such as maize, after the kenaf can be practised to reduce the disease incidence. Annor (1966) reported that the use of crop rotation has been effective in controlling bean anthraenose in bean fields in Australia. Field sanitation which involves the destruction of plant thrash and weed hosts by burning can also be applied. Berger (1977) reported that this can minimize losses due to the disease by reducing initial inoculum through reduction in the number of propagales before a crop is planted.

The finding of roselle as host of the fungus suggests that it can serve as a host during the wet season contributing to the high rate of disease increase on the kenaf since they are generally planted in close proximity on the farmers' fields. It also serve as an alternative host during the dry season and, therefore, acts as a source of primary inoculum that would initiate primary lesions if kenaf is grown in its environment in the subsequent season. Non-cultivation of roselle in the kenaf fields in the wet season and as dry season crop will reduce the inoculum arrival on the kenaf field. Reduction in inoculum arrival is a fundamental practice in controlling many diseases (Berger, 1977).

CHAPTER 10

10. PATHOGENIC VARIATION AMONG ISOLATES OF CONIELLA MUSAIAENSIS

10.1 Introduction

It appears that no information is available on the pathogenic variation in C.musaiaensis. Field observations in the kenaf growing areas showed that variations occurred in the severity of the disease on kenaf cultivars from one place to another and onthe same cultivar within the same farm. A study was, thereforeonducted to investigate the possible existence of pathogenic strains of the fungus.

10.2 Materials and Methods

10.2.1 Selection of host differentials

Since there was no record of any work done on pathogenic variation of the fungus, there were no known standard host differentials for the work. However, nine kenaf cultivars were selected from those available in the germplasm. The nine cultivars selected were three resistant, three moderately susceptible and three very susceptible ones. This selection was based on the reactions inthe field of 57 cultivars.

subsequent to artificial inoculation with isolate 2 (Mokwa isolate of C. musaiaensis ; (see chapter 5) in 1984 and 1986.

The varieties selected and their consistent reaction to attack by isolate 2 under field inoculation in the two years were:

Tianung No.	Resistant
AMC 108	"
A60 - 282	"
CUBA 2032	Moderately susceptible
A60 - 280	" "
BG 58/7	" "
CUBA 108	Susceptible
Kenaf AU-75	"
Bomo Local	"

10.2.2 Collection of diseased specimens

Diseased leaf samples were collected from kenaf growing areas in the southern and northern Guinea savanna zones in 1984 and 1986. Each specimen was put in a paper envelope; labelled and taken to the laboratory. Places of collection of the diseased specimens are given in Table 16. On the whole, 51 specimens were collected, 48 from kenaf plants and three from roselle.

Table 15. Sources of C.musaiaensis isolates used in the study of pathogenic variation in C.musaiaensis

Isolate Number	Date of Collection	Origin/Place of collection	Source Variety of Kenaf
1	August 1985	Mokwa	AMC - 108 Kenaf
2	August 1984	Mokwa	Cuba 108
3	September 1986	Zaria	BZ - 1
4	" "	Zaria	HC - 583
5	" "	Zaria	GCD-98-13
6	" "	Zaria	AC 313
7	" "	Zaria	AU 191
8	" "	Zaria	AU 71
9	" "	Zaria	AU 72
10	" "	Zaria	AU 51
11	" "	Zaria	Roselle
12	" "	Zaria	Roselle
13	" "	Zaria	Roselle
14	" "	Zaria	AU 24
15	" "	Zaria	AU 15
16	" "	Zaria	Local line
"	" "	Keffi	not identified
17	" "	Idu (Abuja)	" "
18	" "	Saminaka	" "

Table 16 (Cont'd)

Isolate Number	Date of Collection	Origin/Place of collection	Source (Variety of Kenaf)
19	September 1986	Lafiya (Benue State)	not identified
20	" "	Sabongida	Cuba 108
21	" "	Sabongida	" "
22	" "	Zaria	" "
23	" "	Sabongida	" "
24	" "	Sabongida	" "
25	" "	Sabongida	" "
26	" "	Gidan Waya	not identified
27	" "	Godogodo	" "
28	" "	Godogodo	" "
29	" "	Godogodo	" "
30	August 1987	Zonkwa	" "
31	" "	Zonkwa	" "
32	" "	Zonkwa	" "
33	" "	Zaria	" "
34	" "	Zaria	" "
35	" "	Zango Kataf	Cuba 108
36	" "	Zango Kataf	" "
37	" "	Zango Kataf	not identified
38	" "	Zango Kataf	" "
39	" "	Zonkwa	" "
40	" "	Kafanchan	" "

Table 16 (Cont'd)

Isolate Number	Date of Collection	Origin/Place of collection	Source (Variety of Kenaf)
41	August 1987	Kafanchan	not identified
42	" "	Kafanchan	" "
43	" "	Soba	" "
44	" "	Soba	" "
45	September 1986	Gidan Waya	" "
46	" "	Gidan Waya	" "
47	" "	Gidan Waya	" "
48	" "	Maraban Sabongida	" "
49	" "	Soba	" "
50	" "	Aliade	" "
51	" "	Aliade	" "

10.2.3 Isolation of the pathogen and preparation of inoculum

Isolation was carried out as described in Section 4.1.2 above, except that monoconidial cultures were not prepared. Subcultures of various isolates were grown on PDAS plates and incubated at room temperatures (25 - 30°C). Five 9-day old agar plate (9-cm diameter) cultures of each isolate were harvested as described below.

Each plate was flooded with 25 ml of sterile distilled water and set aside for 10 minutes before the pycnidiospores were harvested by gently rubbing the surface of the culture with sterilized glass rod. The suspension was transferred to a beaker. Each plate was rinsed with sterile distilled water and the suspension transferred to the same beaker.

The suspension was filtered into a 250 ml conical flask through one layer of muslin cloth. The concentration of the conidial suspension was determined with the aid of a haemocytometer. More sterile distilled water was then added to the flask to bring the spore concentration to about 1×10^5 conidial per ml.

10.2.4 Inoculation of the host differentials

The nine selected kenaf cultivars were raised on flame sterilized soil in 12 x 10 cm plastic pots. Six seeds of each cultivar were sown per pot; seedlings were allowed to for 15 days before they were thinned to four seedlings per pot.

The conidial suspension of each isolate was transferred into a Gallenkamp plastic hand sprayer. Four, 28-day old seedlings of each cultivar were inoculated by spraying to run-off with the conidial suspension of one isolate. Inoculation was carried out between 1700 and 2030 hours as described in Section 4.1.3 above; care being taken to ensure that the plants in different pots did not touch each other. The plants were scored for disease at 10 days after inoculation based on a scale of 1 to 9 described in section 5.3. Due to the large number of isolates, a hand sprayer had to be used more than once. In order not to contaminate isolates with one another, the following method was adopted in cleaning the sprayers. After spraying with the suspension of any isolate, the sprayer was thoroughly washed with two changes of tap water and dipped in hot water (which had previously been boiled and kept in a flask) for one minute, and then cooled in sterile distilled water before it was re-used.

As a result of large number of isolates (51 in all) to be tested on each of the nine kenaf cultivars, a large number of potted plants would ideally have been used at the same time. This number of pots was not readily available. Also, available space for the experiment was limited. There was also the logistical problem of inculating so many plants on the same day. Twelve isolates were, therefore, tested at any one time. At each time, Isolate-2 (Mokwa isloate) was included as a check. Thus, as long as isolate-2 continued to give consistent reactions on the nine cultivars, after each set of inculation, it was assumed that there had been no significant change in environmental conditions.

10.3 Results and Discussion

Forty-eight out of 51 isolates tested were collected from kenaf plants while three were collected from roselle plants (Table 16). The isolates were collected from both the southern and northern Guinea savanna zones. The reactions of the nine selected kenaf cultivars to inoculation with pycnidiospore suspension of each of the isolates (Table 17 and Appendix IV) formed the basis of separating the isolates into six classes (Table 18).

The first class contained eight isolates, viz, isolates 3, 4, 8, 10, 21, 27, 30 and 33. It is the most virulent group, attacking all the cultivars used as host differentials.

Table 17. Interactions between isolates of Coniella musaiaensis and 9 selected cultivars of kenaf in the glasshouse.

Isolate of C.musaiaensis	Tianung No.1	AMC-108	A60-282	CUBA 2032	A60-280	BG-58/7	CUBA 108	Kenaf AU-75	Borno Local	SED ±	LSD
1	1.5	4.0	5.3	7.0	7.5	7.0	7.0	7.5	7.0	0.764	1.58
2	1.0	1.0	1.0	4.0	4.5	3.5	8.0	8.0	7.5	0.597	1.23
3	5.0	6.0	7.5	8.0	7.0	7.0	7.0	7.5	8.5	0.573	1.183
4	5.5	6.5	7.0	7.0	7.0	8.5	8.0	8.5	7.0	0.577	1.19
5	1.0	4.0	6.5	7.0	4.5	7.5	7.0	7.0	7.5	0.569	1.175
6	1.5	4.0	6.5	6.0	5.5	7.5	6.0	8.5	8.0	0.967	1.996
7	1.0	1.0	1.0	1.0	1.0	1.0	1.5	2.5	3.0	0.326	0.672
8	5.0	7.0	6.5	7.0	7.0	7.0	8.5	7.5	7.0	0.869	1.557
9	1.0	3.5	1.0	4.5	4.0	6.5	7.5	7.5	7.0	0.608	1.255
10	3.5	7.0	7.5	7.0	7.5	7.0	7.0	8.0	7.5	0.553	1.141
11	1.0	1.0	1.0	1.0	2.0	1.0	1.5	1.0	1.0	ns	ns
12	2.0	3.5	6.5	6.0	6.5	6.5	6.5	7.5	7.0	0.723	1.493

Table 17 (Cont'd)

Isolate of C.musaiaensis	Tianung No. 1	AMC-108	A60-282	CUBA 2032	A60-280	BG-58/7	CUBA 108	Kenaf AU-75	Borno Local	SED ±	LSD
13	1.0	1.5	4.0	1.3	2.0	2.5	4.5	6.5	8.5	0.913	1.88
14	1.0	2.0	2.0	2.5	3.0	3.0	2.5	7.5	7.0	0.573	1.183
15	1.0	3.5	5.5	6.5	4.5	7.5	7.0	7.5	8.0	0.634	1.310
16	2.5	4.0	5.5	5.0	6.0	6.0	7.5	7.5	7.0	0.638	1.317
17	1.0	1.0	1.0	4.5	3.5	4.0	7.0	7.5	7.0	0.656	1.204
18	1.0	1.0	1.0	1.0	1.0	1.0	1.5	2.0	2.5	0.446	0.921
19	1.0	3.5	1.0	4.5	4.5	6.5	7.0	7.3	7.5	0.799	1.623
20	2.5	4.0	6.0	6.0	5.5	6.0	6.5	7.0	7.0	1.153	2.379
21	3.3	5.5	5.5	6.5	6.5	7.0	7.5	7.0	7.5	1.073	2.216
22	2.5	4.5	6.5	5.5	6.0	6.0	8.0	7.0	7.0	0.752	1.551
23	1.5	2.5	6.5	6.0	5.5	5.0	5.0	7.0	7,0	0.544	1.123
24	1.5	2.5	4.5	5.5	6.0	7.0	7.0	7.0	7.0	0.908	1.874
2	1.0	1.0	1.0	4.5	4.0	4.0	7.5	8.5	8.0	0.567	1.230
25	1.0	4.5	6.0	5.0	3.5	7.0	7.5	7.0	7.0	0.761	1.570

Table 17 (Cont'd)

Isolate of C.musaiaensis	Tianung No. 1	AMC-108	A60-282	CUBA 2032	A60-280	BG-58/7	CUBA 108	Kenaf AU-75	Borno Local	SED ±	LSD
26	2.5	4.5	5.5	6.5	5.0	5.5	7.0	7.0	8.5	0.677	1.397
27	5.5	6.5	5.5	6.0	5.5	7.0	7.0	9.0	7.5	1.072	2.211
28	2.0	1.5	4.5	6.0	5.0	7.5	7.0	7.5	7.0	0.805	1.661
29	1.0	4.0	5.0	5.0	4.5	7.0	7.5	7.5	8.0	0.814	1.679
30	3.3	7.0	7.5	5.5	7.0	7.5	7.5	7.0	7.0	0.788	1.625
31	1.0	3.5	1.0	4.5	4.0	5.0	7.0	7.5	7.0	0.627	1.295
32	1.5	2.0	4.5	6.0	5.5	7.0	7.5	7.0	7.5	0.833	1.719
33	5.0	5.5	7.5	7.0	7.0	7.0	7.5	7.0	7.0	0.649	1.339
34	1.5	4.5	5.5	5.0	5.5	6.0	7.0	7.0	7.0	0.733	1.513
35	1.0	1.0	1.0	1.0	1.0	1.0	1.5	1.0	1.5	ns	ns
36	1.5	3.0	3.5	5.5	6.0	7.0	7.5	7.5	7.5	1.027	2,12
2	1.0	1.0	1.0	3.5	3.5	4.5	8.5	8.5	8.5	0.582	1.24
37	1.0	3.0	2.0	2.5	2.0	3.0	4.5	6.0	6.5	0.945	1.951
38	1.0	3.5	1.0	4.0	3.5	5.5	7.0	7.0	8.0	0.704	1.452

Table 17 (Cont'd)

Isolate of C.musaiaensis	Tianung No. 1	AMC-108	A60-282	CUBA 2032	A60-280	BG-58/7	CUBA 108	Kenaf AU-75	Borno Local	SED ± SE	LSD
39	1.0	2.0	2.0	2.0	3.0	2.5	3.5	5.0	5.5	0.842	1.737
40	2.0	4.5	5.5	7.0	7.0	7.0	7.5	7.0	7.0	0.793	1.638
41	1.0	1.0	1.0	4.0	4.5	4.5	7.0	7.5	7.5	0.701	1.446
42	1.0	1.0	1.0	1.0	1.0	1.5	1.0	1.0	1.0	0.236	ns
43	2.0	1.5	3.5	5.5	6.0	7.0	8.0	7.5	7.5	0.638	1.317
44	1.0	1.5	2.5	2.0	3.0	3.5	4.5	7.5	7.5	0.755	1.557
45	1.0	3.5	1.0	3.5	3.5	5.5	7.5	7.0	7.0	0.518	1.070
46	1.0	2.0	3.0	2.5	3.5	2.5	4.0	5.0	5.0	0.612	1.264
47	1.0	3.0	2.5	2.5	3.0	4.0	2.0	7.0	7.0	0.791	1.632
48	1.0	2.0	1.5	2.5	1.5	2.0	4.5	6.0	6.0	1.041	2.148
49	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	ns	ns
50	1.0	3.5	1.0	4.0	4.5	5.0	7.5	7.0	7.5	0.527	1.087
51	1.0	2.5	2.0	3.0	3.0	2.5	2.0	7.0	7.0	0.612	1.264

Table 18. Pathogenic groups of isolates of C.musaiaensis on nine kenaf cultivars in the glasshouse.

Reactions class of C.musaiaensis	Response of Kenaf cultivars to isolates									Examples of isolates
	Tianung No. 1	AMC-108	A60-282	CUBA 2032	A60-280	BG-58/7	CUBA 108	Kenaf AU-75	Bomo Local	
1	MR/MS	MS/S	MS/S	MS/S	MS/S	S	S	S	S	33, 3, 4, 8, 10, 21, 27, 30.
2	MR	MR	MS	MS/S	MS/S	MS/S	S	S	S	1, 6, 12, 16, 20, 22, 23, 26, 34, 40, 5, 15, 25, 29.
3	HR	HR	HR	MS	MS	S	S	S	S	32, 24, 28, 36, 43.
4	I	MR	I	MR	MR	MS	S	S	S	31, 38, 45, 50, 9, 19.
5	I	HR	HR	HR	HR	HR	HR	S	S	14, 47, 51, 13, 37, 39, 44, 46, and 48.
6	I	I	I	HR/MR	HR/MR	MR	S	S	S	2, 17, 41.

KEY:
I = Immune
HR = High resistance
MR = Moderately resistant
MS = Moderately Susceptible
S = Susceptible

Five of these isolates were collected from Zaria in the northern Guinea savanna while the others were collected from the southern Guinea zone.

The second group contained fourteen isolates (1, 6, 12, 16, 20, 22, 23, 26, 34, 5, 15, 25, 29 and 40). It contained the largest number of isolates. Isolates in this group attacked all cultivars except Tianung No. 1 which gave a high level of resistance (Shimony and Friend, 1975). Among this group is isolate 12, obtained from roselle in Zaria. The 14 isolates were obtained from Zaria, Zonkwa, Kafanchan, Sabongida, Godogodo and Mokwa, the areas where kenaf has been under cultivation for many years. The third class contained five isolates (namely, 24, 28, 32, 36 and 43). These isolates were pathogenic on six of the kenaf differentials. They induced high levels of resistance on the three cultivars that earlier were considered to be resistant to the pathogen.

The fourth group contained six isolates. These are isolates 9, 19, 38, 31, 45 and 50. These isolates were pathogenic on seven out of the nine differentials and non--pathogenic on cultivars Tianung No. 1 and A60-282.

The fifth class contained nine isolates (13, 37, 39, 44, 46, 48, 14, 51, and 47) which were pathogenic on two

cultivars (Kenaf AU-75 and Bomo Local) and non-pathogenic on the other seven. Among this class was isolate 13 obtained from roselle. While Tianung No.1 was completely immune to attackby isolates in this group the other cultivars either exhibited high resistance or moderate resistance.

The sixth group contained three isolates (2, 17 and 41). Three each of the nine kenaf differentials showed resistant, moderately resistant and susceptible reactions, respectively to these isolates. The remaining six isolates - isolates 7, 11, 18, 35, 42 and 49 - were non-pathogenic on the host differentials. Although the fungus could be found attacking the stem in extreme cases, it essentially induced leaf symptoms in kenaf plants. The groups are based on leaf lesions and the host reactions have been classified into the following classes - Immune (I) highly resistant (HR), moderately resistant (MR), moderately susceptible (MS) and susceptibel (S) - as described in Section 6.1.1. A cultivar which showed no visible symptom of the disease was considered to be Immune (I).

The sites of collection of isolates in different pathogenic groups are given in Table 16 which was compiled to find out if any pathogenic groupings were associated with sites of collection of the materials from which the isolates were obtained. Table 16 shows the pathogenic class of an

isolate was not location specific since isolates from the same location were found in different pathogenic groups.

The three isolates obtained from roselle fell within three different pathogenic group indicating the host species probably had little effect on the pathogenic group of the isolates. Since the reactions of kenaf lines to the isolates ranged from complete immunity and full susceptibility, depending on the pathogenicity of the isolate and susceptibility of the host cultivar, it appears that in Nigeria C.musaiaensis populations with different levels of pathogenicity in kenaf exist. Also isolates from kenaf could attack roselle and vice versa.

Agar plate culture of four isolates (5, 14, 17 and 34) were slightly different in colour (appearing whitish) compared to the other isolates. However, these four isolates were found to fall into four different pathogenic groups, suggesting that differences in colour of colonies were not associated with differences in pathogenicity. This observation agreed with earlier one by Ozolua (1981) and Emechebe (1986) who reported no relationship between cultural characteristics and pathogenicity of different isolates of Colletotrichum graminicola in sorghum and C.capsici in cowpea, respectively. Since the major differences between the isolates of Coniella leaf spot pathogen were in their interactions under glasshouse

conditions with nine kenaf cultivars the six pathogenic groups could be designated.

The inclusion of isolates from different ecological locations in the same pathogenic group indicates the widespread occurence of the isolates with divergent pathogenic potentials in the kenaf growing areas of the northern and southern Guinea savanna of Nigeria. The susceptibility of Tianung No.1 to only 7 out of 51 isolates of the pathogen suggests that it possesses genes for broad-based resistance to the pathogen. This makes it the best of all the lines that could be used in developing kenaf cultivars with horizontal resistance (sensu Van der Plank, 1963) to the pathogen.

SUMMARY AND CONCLUSIONS

The work reported in this thesis has shown that a leaf spot disease of kenaf in parts of northern and southern Guinea savanna zones of Nigeria is induced by Coniella musaiaensis B.Sutton var hibisci B. Sutton. This conclusion is based on frequent association between the fungus and the symptoms of the disease, isolation of the fungus from lesions, study of morphological characteristics of the fungus, pathogenicity tests and the subsequent confirmation of the identity of the organism by the Commonwealth Agricultural Bureau, International Mycological Institute (CMI), London, England.

Field observations showed that Coniella leaf spot usually starts at the leaf margin (although it could start in any part of the leaf surface) as water soaked spot which expands in the direction of the leaf petiole. Field survey for the incidence and severity of the leaf spot in the southern Guinea savanna of Nigeria showed that the pathogen attack kenaf leaves in all farms visited. Surveys conducted in September 1984 showed that the incidence ranged between 41.5 and 98.2 percent while the severity based on a visual scale of 1 - 9 ranged from 4.6 to 8.6.

Evaluation of 57 kenaf cultivars for their reactions to Coniella leaf spot under field conditions in 1984 and 1986 revealed that only three cultivars, namely, Tianung No.1, A60-282 and AMC-108 Kenaf, were consistantly resistant. However, when the 57 cultivars were screened under glasshouse conditions, only Tianung No.1 showed complete immunity while cultivars AMC-108 kenaf, A60-282 and Local line 1 exhibited high resistance. These four cultivars could be useful as sources of resistance in developing kenaf varieties resistant to the disease. Seven out of the 57 cultivars exhibited moderate resistance while 21 and 24 cultivars were moderately susceptible and susceptible respectively in the glasshouse.

When kenaf was planted at fortnightly intervals as from first week of May up to early August, it was observed that May- and June-sown crops escaped severe disease attack, produced significantly (P= 0.05) taller plants that gave significantly (P = 0.05) higher fibre and grain yields when compared with July- or August-sown crops. It appears that July- and August-sown crops were exposed to high relative humidity and heavy rainfall accompanied with strong winds which favoured inoculum production and dispersal as well as infection of late-sown crops at younger stages of their growth and development than earlier-sown crops which also provided

for infection of later-sown crops. Such late-planted crops also flowered at younger ages in response to favourable photoperiod later in the season. They, therefore, produced low fibre and grain yields. In Mokwa area of the southern Guinea savanna of Nigeria, early planting in May and June, therefore, appears to be preferred to late planting in July and August.

Application of combination of N, P and K fertilizers as well as application of N alone encouraged vigorous growth of kenaf but appeared to predispose the crop to severe attack by the Coniella leaf spot resulting in a reduction in grain yield when compared with plots which received combined N and P or N and K. Also application of N and K significantly ($P = 0.05$) delayed the number of days to the appearance of the first symptoms of the disease; it also reduced significantly Coniella leaf spot severity with concomitant increase in grain yields when compared with plots that received only P or N or K. However, the combined application of N, P and K as fertilizer resulted in more disease attack in both years but did not lead to significant reduction in fibre yields. The implication of these results is that NPK, compound fertilizer which enhanced leaf spot development with no comparative reduction in fibre yield, could be used as fertilizer if the ultimate aim of planting kenaf

fertilizer if the ultimate aim of planting kenaf is for fibre, while combinations of N and P or N and K which significantly delayed the appearance of the disease symptom and significantly increased grain yield could be recommended for use if grain yield is required.

A laboratory study has shown that carbendazim completely inhibited the mycelial growth of C.musaiaensis in PDA, even at concentration as low as 12.5% of the manufacturer's recommended dosage. Benomyl was also equally effective, while Calixin M and mancozeb were less effective, and effected complete inhibition of mycolial growth of the fungus only at twice their recommended rates.

Evaluation of foliar sprays of the four fungicides showed that they gave effective control of the disease when they were applied at 2-weekly intervals compared with the unsprayed check. However, application of sprays of mancozeb, carbendazim and Calixin M at 7-day to 28-day intervals in both 1984 and 1986 did not produce significant differences in fibre yields among different spray regimes, including the unsprayed check.

In contrast, application of benomyl sprays at 7-, 14- and 21-day intervals in 1984 resulted in higher fibre yields compared with yield of unsprayed check plots. However, in 1986, the plots given the above benomyl sprays did not produce higher

fibre yields than the unsprayed check plots. It appears that control of the Coniella leaf spot of kenaf by foliar application of fungicides has little or no advantage in terms of increased fibre yields. This implies that if the ultimate aim of planting kenaf is for fibre production there may be no need to apply fungicide sprays.

Host range studies showed that roselle (Hibiscus sabdariffa L.), cotton (Gossypium hirsitum L.), okra (Abelmoschus esculentus L.), and pepper (Capsicum annum.L.) were susceptible to C.musaiaensis and could serve as its alternative hosts. Of these, typical symptoms were produced in cotton, roselle and okra which belong to the family Malvaceae as does kenaf. The symptoms in pepper (a solanaceous plant) were atypical, resembling those associated with hypersensitive response. Among the malvaceous hosts, roselle was the most susceptible followed by okra and then by cotton. Cotton and okra as well as pepper are being reported for the first time as alternative hosts of C.musaiaensis.

Pathogenic variation in 51 isolates of C.musaiaensis collected from the southern and northern Guinea savanna zones of Nigeria were studied by artificial inoculation of nine kenaf cultivars under glasshouse conditions. The results indicated the existence of six pathogenic groups which were

based on the response of the nine selected cultivars to inoculation by each of the isolates. The distribution of the pathogenic groups did not appear to be related to the geographic location and ecological features of the sites from which the isolates were obtained. Hence they were not location specific. However, more pathogenic groups were found in areas where kenaf is cultivated in a large scale. This is the first record of pathogenic variations in isolates of *C.musaiaensis*. The most resistant cultivar, Tianung No.1, possibly possessed genes for broad based resistance to the pathogen and could serve as a good material for breeding for resistance to the Coniella pathogen.

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APPENDIX 1

Identification Services

Commonwealth Mycological Institute

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Surrey
England TW9 3AF
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21 November 1983

Report No. H1016/83/YN3 & H1017/83/YN3

Your reference

Report on Cultures sent by:
Dr. A.A. Adeoti
Plant Pathologist
Mokwa Agricultural Research Station
Ahmadu Bello University
P.M.B. 101
Mokwa via Jebba
Nigeria

Specimen number	Herb.IMI number	Identification
		Report from Dr. E. Punithalingam
K ✓ Isol. ex Hibiscus annabinus	280998 } 280999b }	Coniella musaiaensis B. Sutton var. hibisci B. Sutton ✓
		Report from Dr. A. Sivanesan
Isol. ex Hibiscus annabinus	280999a	Curvularia verruculosa Tandon & Bilgrami ex M.B. Ellis state of Cochliobolus verruculosus (Tsuda & Ueyama) Sivan. (discarded).

While all reasonable care is taken to ensure the accuracy and reliability of an identification report prepared by the Institu
liability can be accepted by the Institute, its members, staff or agents in respect of any loss, damage or injury (whether fr
otherwise), howsoever caused, which may be suffered as a result of the identification.

APPENDIX II

Selected Properties of the soil at the Experimental Site in Mokwa - 1984 and 1986

	1984				1986			
	Soil Depth				Soil Depth			
	0 = 15 cm		15 - 30 cm		0 - 15 cm		15 - 30 cm	
	Before Planting	After Harvesting	Before Planting	After Harvesting	Before Planting	After Harvesting	Before Planting	After Harvesting
Organic C(%)	0.44	0.54	0.38	0.39	0.054	0.65	0.45	0.37
Available P (PPM)	12.05	20.03	4.5	5.0	12.37	33.4	10.03	10.39
Exchangeable K (meg/100 g)	0.038	0.07	0.070	0.09	0.078	0.18	0.09	0.1

APPENDIX III

Mean Temperature and Rainfall at Mokwa - 1984 and 1986

Month	1984		1986	
	Mean Rainfall (mm)	Mean Air Temperature (^{o}C)	Mean Rainfall (mm)	Mean Air Temperature (^{o}C)
January	-	23.5	-	24.9
February	-	24.5	0.08	23.5
March	18.2	26.5	10.0	24.9
April	42.6	28.3	34.9	27.6
May	156.8	26.2	69.8	28.2
June	232.8	25.5	160.9	25.1
July	106.3	21.9	191.3	22.7
August	164.0	21.4	78.5	21.6
September	146.7	20.9	202.1	21.5
October	40.8	22.2	43.1	23.8
November	-	23.5	1.8	24.5
December	-	24.9	-	22.1
TOTAL	908.2		792.48	

APPENDIX IV

Interaction* between isolates of Coniella musaiaensis and selected cultivars of Kenaf in the glasshouse.

C.musaiaensis isolates	Resistant Tianung No.1	Cultivars AMC-108 kenaf	Moderately A60-282	Susceptible CUBA 2032	Susceptible A60-280	Susceptible BG-58/7	Susceptible CUBA 108	Cultivars Kenaf AU-75	Bomo Local
1	1113	3553	5557	7777	7995	7957	7777	7977	7777
2	1111	1111	1111	3355	5535	3335	9779	9977	7779
3	5555	5577	9777	9797	7777	7777	7777	7977	9997
4	5575	7757	7777	7777	7777	9997	9779	7999	7777
5	1111	3355	7757	7777	5553	7977	7777	7777	7777
6	3111	3553	5777	7935	7555	7797	5757	9799	7997
7	1111	1111	1111	1111	1111	1111	1311	1333	3333
8	5555	7777	7757	7777	7795	7777	9757	5997	5757
9	1111	3335	1111	5553	3535	7775	7977	7779	7777
10	3335	7777	7977	7777	7797	7777	7777	9977	7977
11	1111	1111	1111	1111	1331	1111	1311	1111	1111
12	3311	3335	5777	5577	7757	5777	7555	7797	7777
2	1111	1111	1111	5335	3553	5535	9977	7779	9977
13	1111	1311	3311	1311	3113	1333	3555	7577	9753
14	1111	3311	1133	1333	3333	3333	3133	7797	7777
15	1111	3335	7555	7775	3555	7977	7777	7977	9977
16	3313	5533	7555	5555	7755	7575	7779	7977	7777
17	1111	1111	1111	3555	3335	5533	7777	7797	9775
18	1111	1111	1111	1111	1111	1111	3111	3311	1333
19	1111	3335	1111	3555	5535	7577	7977	7795	9757
20	3313	5533	7593	7755	7555	3759	7757	7777	5797
21	5333	7555	5539	7757	5777	5977	9975	7957	7797
22	1333	5553	7757	7775	5575	5757	7979	7777	5797
23	3111	5333	7577	7755	7555	5555	5555	7777	9757
24	3111	3331	5535	7555	3579	7777	5797	7777	7777

APPENDIX IV CONT'D.

C. musaiaensis Isolates	Cultivars Tianung No.1	AMC-108 kenaf	Moderately Susceptible A60-282	CUBA 2032	A60-280	BA-58/7	Susceptible Cultivars CUBA 108	Kenaf AU-75	Bomo Local
2	1111	1111	1111	5553	3553	5353	7977	9799	7799
25	1111	3555	7575	5555	3335	7777	5799	7777	9775
26	3313	5553	7555	7775	5555	5755	7777	7597	9997
27	9535	7577	5755	5397	5755	7777	7597	9999	7797
28	3113	1113	5535	5757	5555	7977	7759	7995	7777
29	1111	5533	5555	7553	5355	7777	7977	7599	7799
30	3533	7777	7797	7555	7777	7977	5997	7777	7579
31	1111	5333	1111	3555	3553	5555	7777	5799	7777
32	1131	1133	3555	5397	5557	7777	7977	7777	9777
33	5555	5755	7977	7777	7975	7777	9957	7957	7777
34	1311	5535	5755	5555	7555	9537	7777	7777	7777
35	1111	1111	1111	1111	1111	1111	1311	1111	3111
36	1113	3333	3533	5575	7593	7777	7977	9577	7959
2	1111	1111	1111	3353	3533	5553	7999	9799	9997
37	1111	3333	3113	3331	1133	3333	5535	7953	7559
38	1111	3353	1111	5533	3533	7555	7579	7777	9797
39	1133	3113	3311	1133	3333	3133	5333	5555	5593
40	1111	3555	7555	7777	7957	7777	7797	7777	7959
41	1111	1111	1111	3535	5553	3537	7777	9777	7797
42	3311	1111	1111	1111	1111	3111	1111	1111	1111
43	1111	1311	5333	5755	5757	7777	7997	7977	9777
44	1111	1113	3331	3311	3333	3533	5553	7977	5799
45	1111	3335	1111	3335	3335	7555	7977	7777	7777
46	1111	3113	3333	3313	3553	3331	3355	5555	5555
47	1111	3333	3133	3331	3333	3553	3113	7957	9957
48	1111	1133	1311	1333	1113	3113	5355	3975	5397
49	1111	1111	1111	1111	1111	1111	1111	1111	1111
50	1111	3353	1111	5533	5553	5555	9777	7777	7977
51	1111	3331	3113	3333	3333	3331	3113	7777	9757

* Each number represents the disease severity score on a plant.

APPENDIX V

Brief description of Coniella species

Sutton (1980) has described Coniella species as being characterised by hyaline, immersed mycelium, which are branched and septate. The pycnidia are globose, pale brown, immersed to semi-immersed, unilocular and thin-walled. The ostiole is simple, circular, central and not protruding. The conidiophores are usually absent.

The conidia in Coniella species could be olivaceous to brown, base truncate apex to obtuse to almost apiculate, smooth and thin or thick-walled. The conidia of Coniella musaiaensis var hibisci is however hyaline, thin-walled, fussiform and slightly curved. The sizes vary from 11mμ to 16mμ in length and 3.5mμ to 5mμ in breadth. This organism has been found causing large spreading necrotic lesions on Hibiscus species usually in Nigeria.