DISSEMINATION OF CASSAVA BACTERIAL BLIGHT (CBB) *Xanthomonas campestris* pv. *manihotis* (Arthaud-Berthet and Starr) Dye BY INSECTS

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DECLARATION

I, G.W. Otim-Nape, hereby declare that this thesis and the work therein is my own original work and has not been submitted for a degree in any other University.

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DEDICATION

This work is dedicated to my parents, Jesika and Joseph Ogwal; to my wife Joyce and my children Jeffrey, Gerald, Annette and Ivan.

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TABLE OF CONTENTS

ACKNOWLEDGEMENTS vi

LIST OF TABLES viii

LIST OF FIGURES ix

LIST OF APPENDICES x

ABSTRACT xi

INTRODUCTION 1

The Problem 1

Objectives 5

Approaches and Thesis Organisation 6

REVIEW OF LITERATURE 7

History and Geographical Distribution . . 7

Casual Agent 9

Host Range 12

Epidemiology 13

Insect Transmission of Bacterial Plant Pathogens 16

MATERIALS AND METHODS 23
Experimental Site 23
Experimental varieties 25
Experimental Layout 27
Statistical Procedures 41

RESULTS 44
Insects Associated with CBB 44
Possible Dissemination of Cassava Bacterial Blight by Insects 56
Potency of Bacterial Isolates from Insects 62

DISCUSSION 71
Insects Associated with CBB 71
Possible Dissemination of Bacterial Blight by Insects 81
Potency of Bacterial Isolates from Insects 84

SUMMARY AND CONCLUSION 89

REFERENCES CITED 93

APPENDICES 129

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LIST OF TABLES

		Page
Table 1.	Cultural and Biochemical characteristics of Xanthomonas campestris pv. manihotis	11
Table 2	Characteristic Soil Profile in Experimental Fields	24
Table 3	Insects species and Numbers on Bacterial Blight Infected and on healthy cassava leaves	45
Table 4	Insect species and numbers on Bacterial Blight Infected cassava leaves at three different times of day	48
Table 5	Mean monthly Numbers of Insects on Bacterial Blight Infected cassava leaves	52
Table 6	Insect Behaviour on Bacterial Blight Infected cassava leaves	54
Table 7	Mean Insect Population visiting cassava leaves after the application of mollasses and Brewers yeasts on cassava at different times of 1986/87.....	57
Table 8	Insect Feeding Sites on Bacterial Blight Lesions after the application of Dimethoate, Mollasses and Brewers Yeast on cassava at different times of 1986/87	58
Table 9	Mean Bacterial Blight Lesions on Cassava Leaves after Application of Dimethoate, Mollasses and Brewers Yeast on cassava at Different Times of 1986/87	59
Table 10	Mean colonies ($x10^3$) of *X. campestris* pv. *manihotis* isolated from the body surface and faeces of insects associated with CBB lesions	63
Table 11	Mean Number of Days to CBB symptoms Induction by Isolates of *X. campestris* pv. *manihotis* from body surfaces and feaces of Insects Associated with CBB lesions	69

LIST OF FIGURES

		Page
Fig. 1	Field layout of experiment 2	32
Fig.2	Inoculation of cassava leaf lobes with a suspension of bacterial isolates from insects	38
Fig. 3	Inoculated cassava leaves are covered with transparent polythene bags for 24 hours	38
Fig. 4	The system used for feeding insects with bacterial blight infected leaves	40
Fig. 5	The system used for collecting feaces from insects	40
Fig. 6	Total number of insects on healthy and on bacterial blight infected cassava at different dates	46
Fig. 7	Total number of insects on healthy and on bacterial blight infected cassava leaves of four varieties	46
Fig. 8	Mean number of each of the twenty insect species on bacterial blight infected cassava leaves at 6.30–7.30 a.m., 12.30–1.30 p.m. and 5.30–6.30p.m. of selected months of 1985/86	49 & 50
Fig. 9	Symptoms of cassava bacterial blight after inoculation of cassava leaves with isolates from insects	66
Fig. 10a	Scalding of cassava leaves after five days after inoculation of leaves with isolates from body surface of Dermapteran	68
Fig. 10b	Scalding and tattering of cassava leaves after inoculation of the leaves with isolates from body surface of Dermapteran – 10 days after inoculation.....	68
Fig 11a and b	Recorded weather data during the experimental periods 1985 and 1986	77

LIST OF APPENDICES

Appendix 1. Pictorial Presentation of Insects Associated with CBB 127

Appendix 2 Order, Family, Genera and Species of Insects Associated with Bacterial Blight Infected cassava leaves 128

Appendix 3 Estimated % of Bacterial Blight Infection 129

ABSTRACT

Bacterial blight of cassava, Xanthomonas campestris pv. manihotis (Arthaud-Berthet, Starr) Dye, is one of the most important diseases of cassava in Uganda. The infection of cassava by the disease may result in defoliation, tip dieback, and reduction in tuber yield of up to 90-100% in susceptible varieties.

This thesis reports results of three field, laboratory and green house studies conducted during 1985-87 on the role of insects in the epidemiology of cassava bacterial blight. The first experiment was carried out to identify the most suitable cassava varieties and the insects associated with them. Four cassava varieties (Bukalasa 11, Bukalasa 8, Bintiminti and Ebwanateraka) were used. The insects associated with healthy and CBB infected leaves were monitored monthly. The monthly records were taken at early morning, mid-day and evening. The second experiment was carried out to prove that the insects associate with CBB infected leaves and to establish the role they play in CBB epidemiology. Mollasses and brewers' yeasts were used to mimic insects

from CBB infected to CBB free plants while dimethoate and unsprayed control were used to exclude insects from visiting healthy cassava and to represent natural conditions respectively. The last experiment was carried out to ensure the potency of bacterial isolates from insects. Bacterial isolates from body surfaces and faeces of the insects collected from the field were cultured in the laboratory. Their population in culture and infectivity in cassava seedlings were determined.

A total of twenty insect species from seven orders were associated with bacterial blight infected cassava plants. The insects preferred bacterial blight infected than healthy cassava leaves. Neither the cassava varieties used nor the time of the year affected the insect preference.

Nevertheless, time of the day significantly affected the number and abundance of each insect species on the infected cassava leaves. All the insect species studied were most abundant on the infected leaves during the warmer part of the day (12.30 - 1.30 p.m.) than either

earlier in the morning (6.30 - 7.30 a.m.) or late in the afternoon (5.30 - 6.30 p.m.).

The population of insects on the infected cassava leaves reached two peaks. Variations in peak populations were probably due to the age of the crop, the number of bacterial blight lesions on the crop and the weather factors (particularly rainfall).

All the insect species studied had movements directed towards locating bacterial blight lesions and were probably mediated by some kariomones from blighted lesions. Once a lesion was located, the insect either sucks the exudates from the lesion or chews parts of the lesion. The feeding behaviour and sites of chewing species were of two types. For instance Catantops momboensis centralis and Lagria villosa preferred feeding on edges of the leaf blight lesions on which they create irregular feeding sites. Systates castaneipennis prefers to feed on the middle of blight lesions and creates smooth round "holes".

Mollasses and brewers' yeast sprayed on to healthy leaves, significantly attracted higher number of insects to the crop. Insect feeding sites and bacterial blight lesions were also significantly higher in plots treated with the 'attractants'. The number of lesions were highly correlated with the number of insects: implying that the insects were responsible for the observed increases.

The application of Dimethoate to suppress insects on cassava, resulted in a significant reduction in the number of bacterial blight lesions on cassava and in the insect feeding sites. In general, it was estimated that under natural conditions, the insects contributed up to 35.43% of bacterial blight epidemiology. When mollasses and brewers yeasts were used, the insects contributed 41.19%-56.64% of bacterial blight increase in the field.

Generally the population of the pathogen isolated from the exoskeleton and feaces of the insects were lower than those from bacterial blight infected lesions. Cassava leaves inoculated with isolates produced typical bacterial blight lesions.

The senario of CBB epidemiology seems intricably linked up with food ecology of many insect species particularly Coleopterans and Dipterans. The insects are attracted to bacterial blight infected cassava leaves by kariomones from disintegrating cassava leaf tissues. They acquire the pathogen while feeding on bacterial blight lesions and disseminate them to healthy cassava plants as they move about within a cassava field. The insects play an important role in bacterial blight epidemiology and measures aimed at controlling the disease should integrate the use of insecticides in reducing the number of insects.

CHAPTER I

INTRODUCTION

1.1. The Problem

Cassava, Manihot esculenta Crantz is one of the most important food crops in Uganda (Otim-Nape and Ocitti p'Obwoya 1989). The crop is grown by peasant farmers throughout the country, but the main areas of production are the Eastern, Northern and North Western Uganda (Otim-Nape 1981). The crop is grown mainly for human consumption although small quantities are processed into industrial starch, enguli and animal feed (Otim-Nape 1989). The production of the crop in the country has been increasing. For instance, it increased from 1.92 million metric tonnes in 1970 to 3.9 million metric tonnes by 1985 (Otim-Nape 1989). The factors contributing to the increase in production of the crop are unknown. However, it is likely that the increases are due to expansion in area planted with the crop and not necessarilly due to increases in yield per unit area.

Cock (1981) concluded that any factor which may reduce the leaf life, photosynthetic rate or severely

damages the stems or gives high percentage of early plant death, may severely reduce yields of the crop. He pointed out that pests and diseases may affect yield by destroying plant apices, leaves and storage roots. More than thirty five pests and diseases have been reported on the crop (Lozano and Van Schoonhoven 1975). In some cases these agents cause considerable losses and at times represent a potential danger to the crop and its production.

The diseases and pests that infect cassava have been well reviewed by Lozano and Booth (1975) and by Therberg et al (1987). In Africa, bacterial blight (CBB) Xanthomonas campestris pv. manihotis (Arthaud-Berthet and Starr) Dye is one of the most serious diseases of cassava (Terry 1978). This disease was first recorded in Uganda in Terego county, North Western Uganda (Otim-Nape 1976). It is widespread in Eastern, Northern, North Western and parts of Western Uganda where on poor soils, 90-100% crop loss have been observed on susceptible varieties (Otim-Nape and Sengooba 1981).

In an attempt to contain the disease within the already infected areas, statutory instrument No.27; 1977 prohibited movement of cassava products and vegetative materials from neighbouring countries and between provinces within the country but with little success. The use of tolerant or resistant varieties, bacterial blight free planting material, timely planting to avoid peak rainy season have beeen suggested as possible control measures (Lozano and Sequeria 1974b, Terry 1978, Otim-Nape 1978). Screening for and developing varieties resistant to the disease is being carried out (Anonymous 1986, 1987 and 1988), but this is long term and expensive. Other cultural practices such as use of clean planting material, timely planting and use of optimum spacing have been tried (Anonymous 1986, 1987, 1988). Their effects are minimal.

Detailed epidemiological studies to clarify how environmental factors influence development of the disease have been suggested (Maraite and Meyer 1975). Insects have also been suggested as a possible factor in the epidemiology of the disease (Amaral 1945). In Zaire, infection of cassava by bacterial blight starts from

wounds or insect punctures made principally by Pseudotheraptus devastans Distant (Maraite and Meyer 1975). Large populations of Diptera particularly those belonging to the family Muscidae, have been observed colonising infected plants in Uganda (Nyiira and Otim-Nape 1978). It is therefore likely that insects may play a role in the dissemination of the disease. Maraite and Meyer (1975) suggested that the role of insects in the epidemiology of bacterial blight should be elucidated.

Despite the observations and suggestions, no detailed studies have been carried out to clarify the role of insects in the epidemiology of bacterial blight. Thus, there is no reliable information which pathologist can use as a basis for making a decision on additional control measures for the disease.

1.2. Objectives

The study reported here was conducted with the following objectives:

(i) to identify the species of insects commonly associated with bacterial blight,

(ii) to clarify the role of insects in the epidemiology of the disease,

(iii) to determine how the insects disseminate the blight from diseased to healthy plants in the field and

(iv) to find out whether elimination of the insects which disseminate the blight can help in controlling the spread of the disease in the field.

1.3. Approaches and Thesis Organisation

The results reported in this thesis are from field, laboratory and glass house experiments conducted at Serere Research Station in 1985, 1986 and 1987. Studies on insect species and numbers associated with bacterial blight and those on insect dissemination of the disease were carried out in the field in 1985/86. Isolation of bacterial blight pathogen from insects were carried out in the laboratory while studies on infectivity of the isolates were carried out in the glass house on potted plants in 1986 and 1987.

The thesis is written in six chapters. Chapter 2 is a review of literature, Chapters 3, 4, 5 and 6 are materials and methods, results, discussion and summary respectively. Each chapter is divided into sections and occasionally subsections whenever necessary.

CHAPTER II

REVIEW OF LITERATURE

2.1. History and Geographical Distribution

Bacterial blight of cassava was first reported in Brazil in 1906 and attributed to Bacillus manihotis Athaud-Berthet (Bondar 1912, 1915). More recently the disease has become widespread in South America (CIAT 1971, 1972, 1973; Castano 1972a; Normanha 1971), Asia (Leu and Chen 1972; CIAT 1974) and Africa (Orian 1948; Williams et al. 1973; Persley 1979; Hahn and Williams 1973; Nyango 1978; Otim-Nape 1980; Onyango and Ramos 1978; Butare and Mostade 1978)

Since its first report in 1912, cassava bacterial blight has become increasingly important and in 1940s it was considered the most serious disease of cassava in Brazil (Amaral 1942; Chaeves 1946). Losses caused by this disease on cassava in other South American countries and Asia are also high (Lozano and Sequeria 1974b; CIAT 1973, 1974, 1975; Lue and Chen 1978).

In Africa the disease threatened the crop in Zaire, and was responsible for a widespread famine in that country in the early 1970s (Hahns and Williams 1973). In the former East Central State of Nigeria, losses due to the disease was estimated at Nigerian Naira 24 million (US$38 million) and approximately half of the crop in the area was lost due to the disease (Hahns and Williams 1973; Terry 1974). Serious outbreaks have also been reported in other African countries (Batsima and Mabanza 1976, Terry 1978, Otim-Nape and Sengooba 1977). Terry (1978) concluded that reduction in yield due to the disease varies with the location, level of susceptibility of the varieties, seasonal fluctuations, time of planting, and the magnitude of inoculum density. The reduction in starch content of roots of infected plants, and the destruction of planting material due to dieback of infected cassava stems all add to the considerable loss (Terry 1978).

2.2. Casual Agent

The casual agent of bacterial blight of cassava (CBB) has been reported under various names Bacillus manihot (Arthaud-Berthet), Bacillus manihotus (Arthaud-Berthet), Bacillus manihoti (Berthet and Bondar) and Bacterium manihotus (Drummond and Hipolito) (Burkholder 1942; Bergey 1948, 1974). More recently the pathogen has been grouped under Xanthomonas campestris along with many other plant pathogenic bacteria which are indistinguishable in laboratory tests but differ in plant host reaction (Bergey 1974; Bradbury 1970). It has been accordingly renamed Xanthomonas campestris pv. manihotis (Athaud-Berthet and Starr) Dye (Bradbury 1970). Cultural and biochemical characteristics of the pathogen have been widely studied by Drummond and Hipolito (1941), Amaral (1942), Burkholder (1942) and Lozano and Sequeria (1974a). They are summarised in Table 1. Distinct strains of the pathogen have been reported by Lozano and Sequeria (1974a), Maraite and Meyer (1975) and Maraite and Weyns (1980). Lozano and Sequeria (1974a) suggested that the taxonomic position of the species required further investigation. They suggested that in some

respects, notably its white colour, ability to accumulate polybitahydroxy butyric acid, fast growth, and relatively high optimum temperature, the pathogen is akin to group III of the Pseudomonas plant pathogens (Bergey 1974).

The symptoms of bacterial blight, X. campestris pv. manihots on cassava are characterised by: angular leaf spots especially on the under surface of the lower leaves, leaf blight, wilting, defoliation, tip dieback and gum exudation on petioles and young stems. Vascular discolouration of the petioles, young stems and roots of highly susceptible varieties are common (Williams et al. 1973; Lozano and Sequeira 1974a; Maraite and Meyer 1975). Infected cuttings may fail to germinate while many others may develop wilting and death of the young shoots (Lozano 1975).

Table 1: Cultural and Biochemical Characteristics of *Xanthomonas campestris* pv. *manihotis*

	Drummond and Hipolito (1974)	Amaral (1942)	Burkholder (1942)	Lozano and Sequeira (1974)
Size		0.6-0.9 x 1.6-2	0.35-0.93 x 1.4-2.8	0.32-0.49 x 0.76-2.6
Shape	Rods	Rods	Rods	Rods
Mortality	Single polar flagellum	Single polar flagellum	Single polar flagellum	Single polar flagellum
Gram reaction	Negative	Negative	Negative	Negative
Spore formation		--	--	--
Capsule Formation	--			--
Temperature	Killed at 77°C for 10 minutes			
Minimum °C			5	5
Optimum °C			30	30-32
Maximum °C			38	38
Mass doubting times				32°C-46 min. 30°C-47 min. 28°C-55 min.
Colony type		After 48 hrs 30°C: circular white, raised glistening viscoid colonies	Beef agar: white to hyline mucoid growth	(1) Nutrient agar: white, convex, mucoid colonies (2) Reduces tetrazolium salt
Fluorescence on Kings B. Medium				Negative
Gelatin hydrolysis	+ (3 days)	+(8 days)	+	+
Starch	--	+	-	-
Reduction of litmus milk		+		+
H_2S production		-	+	-
Nitrate reduction	+	-	-	-
Indol Production	-	-	-	-

The bacterium normally penetrates the root via stomatal openings and wounds of epidermal tissues (Pereira and Zagatto 1967; Lozano and Sequeira 1974a). Colombian isolates of X. campestris pv. manihotis multiply in the stomatal cavities and then move progressively into the protoxylem and protophloem of adjacent veinlets four days after infection (Lozano and Sequeria, 1974a). The pathogen invades the vascular strands and disintegrates cells of the palisade layer and adaxial epidermis eight days after infection.

The bacteria move from leaf lesions to the stem via the vascular tissue of the petioles, primarily the xylem (Drummond and Hipolito 1941; Amaral 1942; Lozano and Booth 1975) while roots and older stem tissues are rarely invaded (Lozano and Sequeria 1974b).

2.3 Host Range

X. campestris pv manihotis has only been recorded on species of the genus Manihot (Bergey, 1957). There are no reports of host range studies to test potential alternative hosts of the pathogen.

2.4 Epidemiology

2.4.1 Means of spread

The pathogen moves from one area to another in infected planting material (Amaral 1945, Lozano and Sequeria 1974b). Infected cuttings also disseminate the disease from one growing season to the next (Lozano and Sequeria 1974b). Wind and rain-splash are the most important means of local dispersal (up to 10m) (Lozano and Sequeria, 1974b). Dispersal within a field occurs in the direction of the prevailing winds. Lozano (1972) found a high correlation between rainfall and amount of infection.

Bacterial blight may also spread by the movement of soil during cultural operations or by the use of infected prunning implements (Carneiro 1940; Drummond and Goncalves 1953; Lozano and Sequeria 1974b). A low percentage infection (7%) in a susceptible cultivar was also obtained when a heavy suspension ($5x10^3$ cells/ml) of X. campestris pv. manihotis was added to soil in which cassava was growing (CIAT 1974; Ikotun 1976).

Insects apparently disseminate the bacteria over short distances as mechanical carries, rather than specific vectors (CIAT 1973; Nyiira and Otim-Nape 1978; Daniel et al., 1980). CIAT(1973) found that spraying cassava with insecticide reduced spread of the disease by 10-14%, thus suggesting that insects are much less important than rain-splash. It has been suggested that the movement of men and animals through a crop may spread the disease but this has not been proved (Amaral 1945). The pathogen is also seed borne (Elango and Lozano 1980).

2.4.2 Sources of Infection and Survival.

Infected debris and volunteers from a previously infected crop are important sources of inoculum (Persley 1979). Counts of 3.5 x 10^3cells/g were recorded in Colombia in plant debris on the soil surface. The numbers decreased rapidly with depth of soil (CIAT 1973). X. campestris pv manihotis survives more than seven months in necrosed stem pieces stored at 24°C and 80-87% relative humidity (CIAT 1973; Persley 1979). No bacteria stored in the leaves under the same conditions survived

after five weeks. CIAT (1973) demonstrated that fallow period significantly reduced the incidence of the disease in the field. Soil survival studies with pure cultures of X. campestris pv. manihotis in Colombia showed that the cells survive longer in sterile than in non-sterile soils (CIAT 1974). Optimum pH 6.06-6.50 and low organic matter content of soils favour the survival of the pathogen (CIAT 1974). In Nigeria, X. campestris pv manihotis survives up to fourteen months in pellets on the under-surface of diseased cassava leaves (IITA 1974, Terry 1975).

2.4.3. Effects of environmental factors on disease development

There is little experimental data on the effects of environment on disease development. Lozano (1975) and Lozano and Sequeria (1974b) suggested that dispersal of the disease was favoured by high rainfall and that disease spread stops during dry periods. They added that after inoculation a period of high humidity (12h at 100% RH) is necessary for the infection to be established.

The disease is most damaging on sandy loam soils of low fertility and in fields where cassava had been grown continously for several years without fertilizers (Glaser and Ogbogu 1974; Ezumah and Terry 1974; Maraite and Meyer 1975). The highest incidence of the disease normally occurs at the beginning of the wet season (Lozano and Sequeria 1974b).

Control measures for the disease are difficult although some resistant varieties have recently been developed (Normanha and Pereira 1950; Hahn et al. 1974; IITA 1978, 1979, 1980). Amendments to various cultural practices have been tried but with little success (IITA 1974, 1975, 1978; CIAT 1974, Terry 1977).

2.5. Insect Transmission of Bacterial Plant Pathogens

The involvement of insects in the transmission of bacterial plant pathogens is indisputable (Harrison et al. 1980). At least sixty diseases are known to be transmitted by insects (Harrison et al 1980).

Fire blight Erwinia amylovora (Burrill) Winslow et al. was the first bacterial disease shown to be vectored and transmitted by insects (Waite 1891). Since then, as many as one hundred or more insect species have been shown to be involved, in some way, with the transmission, survival and inoculation of the pathogen into the plants (Harrison et al. 1980). Bees are considered the most important vector of the pathogen during blossoming periods (Waite 1906, Beer 1979, Erumett and Baker 1971). Erwinia amylovora has been cultured from ants, aphids, beetles, flies, thrips, lygus bugs, leaf hoppers and various hemipterans collected from pear blossoms and oozing cankers (Waite 1896, 1906; Thomas and Ark 1934a; Falson 1954; Emmett 1971; Parker 1936; Thygesen et al. 1973; Wilde 1971; Beer 1979; Stahl and Luepschen 1977). Jones (1910, 1911a and b), Tullis (1929), Stewart (1913a), Thygescan (1973), and Stahl and Luepschen (1977) noted that fireblight infections always developed around fresh punctures and injuries made by beetles, codling moths and lygus bugs.

The transmission of blackleg and bacterial soft rot Erwinia carotovora var carotovora and E. carotovora var atroseptica of potatoes by dipterous insects particularly Hylemya platura (Meigen) Hylemya florilega (Zetherstedt) and Drosophilla busckii Coquillet have been reported (Leach 1940, Tamini and Bonfield 1969; Molina et al. 1974; Graham et al. 1976; Harrison et al. 1977; Kloepper 1979; Kloepper et al. 1976; Kloepper et al. 1979; Kloepper et al. 1981; Phillips and Kelman 1981). Likewise larvae of the seed corn margots Hylemya antiqua Meigen Hylemya platura Meigen and those of the black onion fly, Tritoxa flexa Wiedeman and Eumeris strigatus Fallen, have been associated with bacterial decay of the onion bulbs (Tylor 1952; Doane 1953). They have been considered important in the survival of the pathogen Erwinia carotovora var carotovora (Doane 1953; Friends et al. 1959; Brooks 1963; Harrison et al. 1980). Eggs of several insect species, particularly Scoptomyza graminum (Fallen) and Elachiptera costa (Loew) transmitted bacterial soft rot of celery, Erwinia carotovora var carotovora (Jones) Bergey et al when maintained on moist celery leaves (Leach 1927). The destructiveness of the disease during the hot dry season

is believed to be due to the changes in the ovipositional behaviour of the insect (Leach 1927; Oglvie et al. 1934). The striped and spotted cucumber beetles Acacymma vittata (Fabricius) and Diabrotica undecimpunetata howardi Barber (both Coleoptera: Chrysomelidae) are major vectors of curcubit wilt Erwinia tracheifila (Smith) Bergey et al. (Carter 1973, Leach 1964). Erwinia stewartii (Smith) Bergey et al. is also transmitted by flea beetles Chaetocnema pulicaria Welsh, Chaetonema denticulata, Diabrotica undecimpuctata howardii (Barber), H. platura (Meig) and by the wheat wireworms Agriotes mancus (Say) (Rand 1923, Rand and Cash 1924; Poos and Elliot 1936; Ivanoff 1933a and b; Frutchey 1936).

The association and transmission by insects of pineapple fruit and heart rot Erwinia crysenthemi Burkholder et al.; Olive knot Pseudomonas savastanoi (Smith) Stevens; bacterial rot of apples Pseudomonas melophthora Allen and Riker; Southern bacterial blight Pseudomonas solanacearum (Smith) Smith; halo blight of beans Xanthomonas campestris pv phaseoli (Smith) Dowson, X. campestris pv phaseoli var fuscans (Burkholder) Starr and Burkholder and X. campestris pv. phaseoli f.sp.

vignicola; by insects have also been reported (Lim and Lowlings 1977; Petri 1909, 1910; Hellmuth 1956; Tamvrias et al. 1970; Hagen 1966; Fytizas and Tzanakakis 1966a and b; Rey 1969; Harrison et al. 1980; Allen and Riker 1934; Miyazaki 1968; Buddenhagen and Elsesser 1962; Vakili and Baldwin 1966; Buddenhagen and Kelman 1964; Revilla 1967; Kaiser and Vakili 1978).

Many casual insect visitors to the walnut catkins are suspected in the transmission of walnut blight caused by Xanthomonas campestris pv juglandis (Pierce) Bergey et al. The white flies Aleurobolus borodensis M and Trialeurodes vaporariorum West respectively transmit gummosis of sugar cane Xanthomonas campestris pv vasculorum (Cobb) Dowson and leafspot disease of geranium Xanthomonas campestris pv. pelargonii (Brown) Starr and Burkholder (Dye) (Bugbee 1962, Bugbee and Anderson 1963, North 1935, Carter 1973).

2.5.1 Insect Transmission of Cassava Bacterial

Insects have been suggested as possible agents for disseminating cassava bacterial blight. In Nigeria, populations of the variegated grasshopper Zonocerus variegatus were found to be high during the early parts of the rainy season (Terry 1974). These grasshoppers moved around infected cassava plants quite freely and their mouth parts and appendages became contaminated with wet bacterial exudation. Aphids, leafhoppers, other insects and mites have also been suggested as vectors of the pathogen (Okpala 1974; Lozano and Whooley 1974). Large populations of Diptera, particularly those belonging to the family Muscidae have been observed colonising infected plants (Nyiira and Otim-Nape 1978). The most common species observed were Drino spp and Musca spp. The insects sucked the oozing exudates from infected lesions. Daniel et al. (1981) observed and detected by immuno fluorescence, X. campestris pv. manihotis in alimentary canal and feaces of Crysolagria cupriana, Ischnotrachelus sp and Zonocerus variegatus

that had fed on infected cassava leaves of test plants. They suggested that the insects could be disseminators of the pathogen.

CIAT (1973) and Nyiira and Otim-Nape (1979) suggested that the insects apparently disseminate the bacteria over short distances as mechanical carriers rather than specific vectors. Insecticide spray in Colombia reduced the spread of the disease by 10-14%, suggesting that the insects are much less important than rain splash (CIAT 1973). Daniel et al. (1981) concluded that the efficiency with which insects disseminate the pathogen is unknown especially for long distances. They suggested that there was need for more investigations.

CHAPTER III

MATERIALS AND METHODS

3.1 Experimental Site

All the experiments were carried out at Serere Agricultural Research Station, Eastern Uganda, 1°32'N, 33°27'E and 1,140m above sea level. The station represents a larger part of the short grassland zone of Uganda, where cassava is most important

Generally, the soil distribution pattern on the Station follows the topographical features - sandy soils on hill pediments leading down to red soils of heavier texture on remnants of the Tanganyika surface (Ollier and Harrop 1959). The dominant soil types in the experimental fields are "shallow brown sandy loams over quartzite on hill pediments" and its characteristic profile is as indicated in Table 2.

The soils are poor in exchangable bases and are frequently less than 50% saturated. The surface soils are usually poor of organic matter, and deficient in phosphate. Since they contain relatively small amounts

Table 2. Characteristic Soil Profile in Experimental Fields

Depth (cm)	Description
0 - 20	Brown sandy loam with very weak sub-angular blocky structures.
20 - 45	Brown sandy loam almost structure less.
45 - 70	Brown sandy loam and sandy clay with weak sub-angular blocky structures.
70 & over	Rotted quartz schist.

Source: Ollier and Harrop (1959)

of free sesqioxide, the soil probably have a low capacity to fix phosphates and could be expected to respond to phosphatic fertilizers (Ollier and Harrop 1959). The experimental fields were fertilised with 125kg/ha single super phosphate before cropping with sorghum (Sorghum vulgare) the previous season.

The natural vegetation of the area is related to an association of the broadleaved savanna woodland dominated by Butyrospermum parkii, Combretum spp. with Hyparrhenia rufa most abundant in the grass layer. As a result of repeated cultivation, most of the trees in the experimental fields have been removed and natural

regeneration has favoured Hyperreniha filipendula on drier sites and Hyperreniha rufa, Panicum maximum, Imperata cylindrica on sites with deeper or heavier soils.

Mean monthly maximum and minimum air temperatures vary from 27 to 33°C and from 17 to 18°C respectively. The mean annual rainfall is between 1114.3 mm to 1270 mm. It is bimodally distributed with peaks occuring in April to May and August to September. A long dry season occurs from November to March.

3.2 Experimental varieties

The four (4) experimental cassava varieties; (Ebwanateraka, Bintiminti, Bukalasa 8 and Bukalasa 11) used in the study were taken from a multiplication block of disease free materials meant for distribution to farmers. Cuttings for the multiplication block had been obtained from plants that had been raised according to the method of Lozano and Wholey (1974) and were, therefore, free from bacterial blight (CBB) and the African cassava mosaic virus (AMV). The choice of the varieties for the experiments were based on their

popularity in the country and on their relative susceptibility or resistance to cassava bacterial blight. The descriptions of the varieties are as follows:

Ebwanateraka

This is an open pollinated variety of unknown parentage. It is susceptible to bacterial blight (CBB) and the African cassava mosaic virus (AMV) but resistant to the cassava greenspider mite (GCM) Mononycellus tanajoa (Bondar). It is early maturing, very high yielding and has good tuber characteristics.

Bintiminti:

This is a local variety collected from Ngetta - Lira district, during the 1930s. It is resistant to bacterial blight (CBB) and the African cassava mosaic virus (AMV) but is late maturing; gives average tuber yield and has good tuber characteristics.

Bukalasa 8 (Aipin Valencia):

This variety was brought to Uganda in 1941 from Amani, Tanzania. It was brought to Amani from Brazil. It is moderately susceptible to bacterial blight and the African cassava mosaic

virus, but resistant to Bemisia tabaci, vector of the virus. It is moderately susceptible to the cassava green spider mite and is late maturing, gives average tuber yield and has good tuber characteristics.

Bukalasa 11 (F 279):

This variety was brought to Uganda from Amani, Tanzania in 1941. It was brought to Amani from Java. It is resistant to bacterial blight and the African cassava mosaic virus but is susceptible to the cassava green spider mite. It is early maturing, gives average yield, has excellent tuber quality but tubers store poortly in the soil after maturity.

3.3 Experimental Layout

The experimental fields were ploughed twice and disc harrowed once with the DEUTZ-DX 90 tractor. The trial areas were marked and levelled with a hand hoe to achieve land uniformity. All the trials were arranged in a randomised complete block design with replications

varying from four to eight. The remaining non-experimental areas were planted with cassava bulk.

Cassava cuttings about 30 cm long and taken from disease-free matured stems of each variety were planted horizontally into the ground. Details of experimental procedures can be found under each experiment.

All the three experiments were designed either as randomised complete block or split-plot designs with replicates varying from two to eight.

Experiment 1: Insects Associated with Cassava Bacterial Blight (CBB)

Stem cuttings of cassava varieties: Ebwanateraka, Bintiminti, Bukalasa 8 and Bukalasa 11, were each planted in 5m x 5m plots at a spacing of 1m x 1m giving 36 plants per plot or 10,000 plants/ha. The varieties were arranged in a randomised complete block design with eight replicates. One line of the CBB susceptible variety (Ebwanateraka) was planted on 5th June, 1985. The line of the susceptible variety separated and surrounded one replicate of the experiment from another. The susceptible

variety was planted 2m away from the experiment and it acted both as a guard row and as a spreader of bacterial blight to the test varieties. The experimental varieties were planted on 30th June, 1985.

On 30th August, 1985, 0.5 ml of a standard suspension of Xanthomonas campestris pv. manihotis (Arthaud - Berthet) Dye previously isolated from CBB diseased cassava leaves and stems, and incubated for five days at 28 - 30°C on Difco nutrient agar, were leaf infiltrated (Perraux 1977) into the 3rd - 4th and 5th topmost expanded leaves of each plant in the spreader rows. The test varieties were allowed to contract and develop bacterial blight epidemic from spreader rows under natural conditions.

Records of CBB blight lesions, insect feeding sites on blight lesions, the different insect species on both healthy and diseased leaves and insect activities (mobility, feeding and resting) on CBB diseased leaves were taken thrice a day i.e. between 6.30-730 a.m., 12.30-1.30 p.m. and 5.30-6.30 p.m. The first record was taken on 10th November, 1985. Subsequent ones were

repeated on the 10th of every month until 10th May, 1986. The trial was harvested on 30th June, 1986 and records of yields and yield components were taken. Specimens of the insects found associated with bacterial blight lesions were sent to the Commonwealth Agricultural Bureau (CAB), International Institute of Entomology (CIE), London (U.K) for identification.

Experiment 2: Dissemination of Bacterial Blight (CBB) by Insects in the Field

Mollasses and brewers' yeast were used to 'attract' insects from cassava plants infected with bacterial blight to plants free from bacterial blight infection. A broad spectrum insecticide, Dimethoate, was used to keep certain plots free from insects. These acted as control plots. Details of the treatments were as follows:-

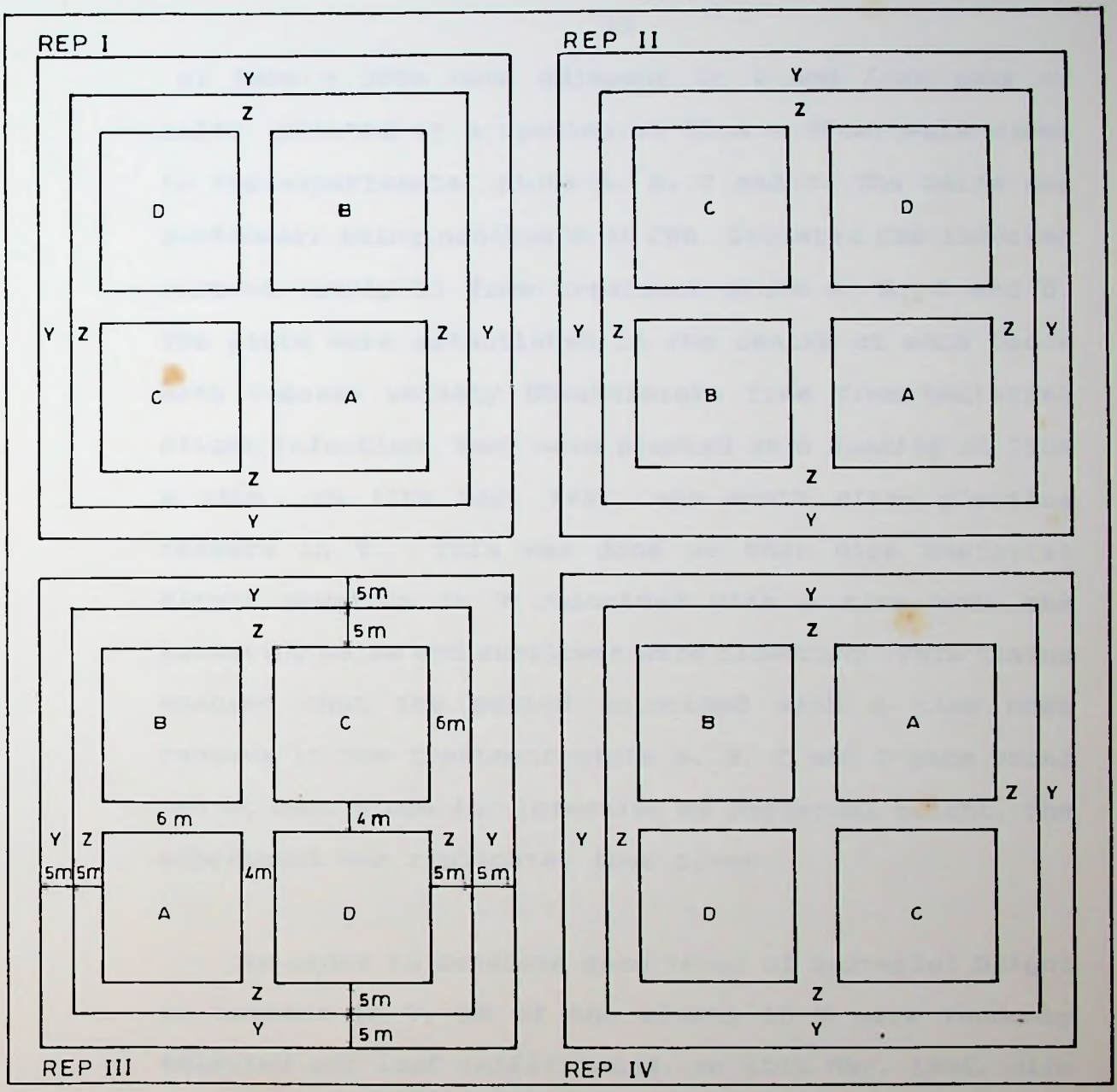
a. Spraying cassava, to drip, with the insecticide in order to keep away insects or mites from the plants; Dimethoate was used at the rate of 1.00 litre/ha,

b. No spraying cassava with insecticide or insect attractant i.e. allowing nature to play its role,

c. Spraying cassava with mollasses to mimic insects from CBB diseased to CBB free cassava; a rate of 2.0 kg/ha was used,

d. Spraying cassava with brewers' yeast to mimic insects from CBB diseased to CBB free cassava; a rate of 2.0 kg/ha was used.

Details of the experimental layout is given in Figure 1. Four blocks, each of 36m x 36m, were marked in the field. Each block represented one replicate. A strip (Y) measuring 5m in width was marked around each block as shown in Figure 1. Cassava was planted on each strip on 11th March, 1986 at a spacing of 75cm x 75cm, using cuttings which were obtained from Ebwanateraka plants severely infected with bacterial blight. This strip was meant to establish high level of bacterial blight infection from where the insects could acquire the pathogen for dissemination to the treatment plots. The strip Y surrounded another strip of 6m (Z) planted, on 11th April, 1986, with closely spaced maize and sunflower. Four rows of sunflower, planted at a spacing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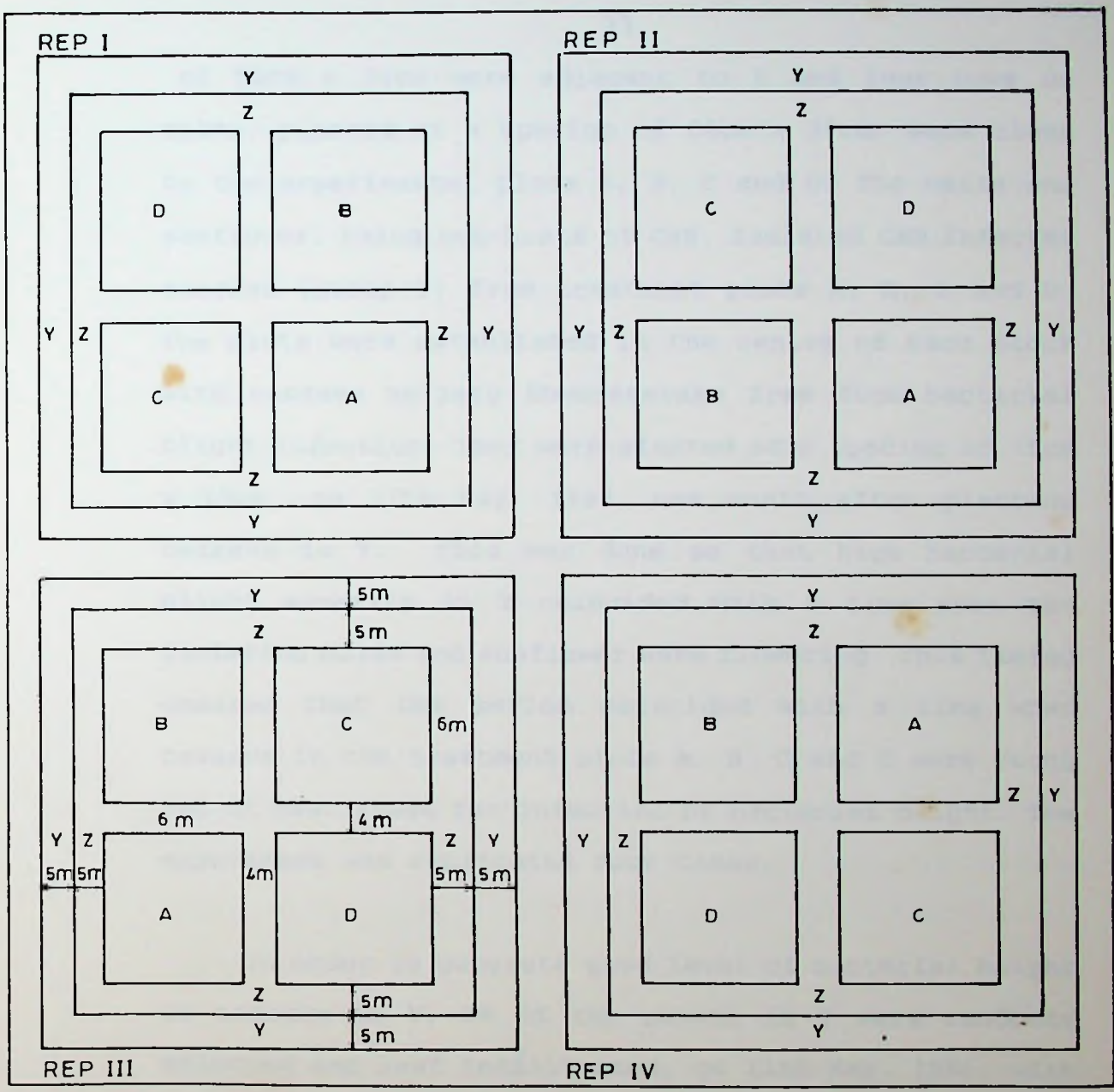
REP I
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Z
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REP II
C
D
B
A
REP III
5m
6m
4m
REP IV

of 50cm x 30cm were adjacent to Y and four rows of maize, planted at a spacing of 60cm x 30cm, were close to the experimental plots A, B, C and D. The maize and sunflower, being non-hosts of CBB, isolated CBB infected cassava (strip Y) from treatment plots A, B, C and D. The plots were established in the centre of each block with cassava variety Ebwanateraka free from bacterial blight infection. They were planted at a spacing of 75cm x 75cm on 11th May, 1986, one month after planting cassava in Y. This was done so that high bacterial blight severity in Y coincided with a time when the isolating maize and sunflower were flowering. This timing ensured that the period coincided with a time when cassava in the treatment plots A, B, C and D were young and at best stage for infection by bacterial blight. The experiment was replicated four times.

In order to generate good level of bacterial blight on cassava in Y, 5% of the plants in Y were randomly selected and leaf infiltrated, on 11th May, 1986, with 0.5 ml of a standard suspension of the bacteria as described earlier in Experiment 1.

As a good level of bacterial blight severity on cassave in strip Y had been reached by 25th November 1986, the isolating maize and sunflower in Z were cut to ground level on that date in order to expose the treatment plots. Treatments A, B, C and D were then imposed. Treatment A (Dimethoate) was always applied 6 hours before applying treatments C (Mollasses) and D (Brewers yeast) as it was feared the smell of Dimethoate would scare away insects from visiting neighbouring treatment plots. The first application of the treatments was made on 25th November,1986, and this was repeated at 10 day interval until 14th January, 1987, the time when record taking on the experiment was terminated.

Records of CBB lesions, insect feeding sites on the lesions and the number of different insect species on cassava leaves infected with bacterial blight were taken every three hours after the insect 'attractants' were applied.

Experiment 3: Potency of Bacterial Isolates from Insects

Insects were collected from diseased cassava leaves on 9th June, 1987 using an aspirator, net or by hand.

The insects collected were: Cantantops momboensis centralis (Orthoptera: Pyrgomorphidae); Nematocerus castaneipennis (Hustache) Coleoptera: Afroeurydemus nigricornis (Selman) Coleoptera: Chrysomelidae; Scymus morelleti Coleoptera: Chrysomelidae: Alticinae; a chloropidae (Diptera); Aspavia albidomaculata (Stal.) Heteroptera: Pentatomidae; Cautires usambarae (Bourgcois) Coleoptera: Hycidae; Podagrice siostedti (Jacoby) Coleoptera: Chrysomelidae: Anticinae; a Foticulinae (Demaptera); Zaprionus sp; Diptera: Drosophilidae; Halydichoris sp. Heteroptera: Pentatomidae; and Lagria villosa (Fabricius) Coleoptera: Lagridae. The remaining eight insects (see Section 4.1.1 and Table 3), previously seen associated with leaves infected with bacterial blight, were difficult to find at the time of collection and were, therefore, not assayed for possible presence of the pathogen on them.

After bringing the insects to the laboratory, they were divided into samples, A and B. Ten insects of each species in sample A were placed in test tubes containing 5ml sterile distilled water and shaken vigorously for 30 minutes. One ml. of the wash water from each tube was

pipetted and serially diluted three times. One tenth (0.1 ml) of the final dilution was pipetted and plated on Nutrient Agar (Lab-Lemco' beef extract 1 gm, yeast extract (Oxoid L20) 2 gm, peptone (Oxoid L37) 5 gm, Sodium Chloride 5 gm, Agar 15 gm, water 1,000 ml pH 7.4) in petri dishes replicated two times. The inoculated petri dishes were incubated at room temperature. Colonies of the bacteria on each petri dish were counted on the third day after inoculation. Colonies of Xanthomonas campestris pv. manihotis were separated from other bacterial contaminants by comparing them with standard colony of Xanthomonas campestris pv. manihotis isolated from cassava bacterial blight lesions and treated as for the insect isolates.

The remaining suspension from each test tube was used in inoculating cassava test plants. One half (0.5 ml) of the suspension was infiltrated into each of six spots per leaf lobe of the second and third top most expanded leaves of 3 months old plants (Figure 2) of cassava variety Ebwanateraka. This was replicated two times. The inoculation was to confirm pathogenicity of the isolates. Each inoculated leaf was immediately

covered with a transparent polythene bag - gauge 250 (Figure 3) in order to maintain high humidity. After 24 hours, the bags were removed. The development of symtoms of bacterial blight at the inoculated points were observed at an interval of 24 hours.

Ten insects of each species in sample B were caged into 250 ml flat bottom flasks into which four CBB blighted cassava leaf lobes were vertically inserted (Figure 4). The top of each flask was covered with a

fixed space

Fig. 2 : Inoculation of cassava leaf lobes with a suspension of bacterial insolates from either body surface or from feaces of insects.

Fig. 3 : The inoculated cassava leaves are covered with transparent polythylene bags for 24 hours.

cheesecloth of varying layers in order to prevent the insects from escaping. This was done immediately the insects were taken to the laboratory and were therefore not starved. The insects were allowed to feed on the leaf lobes while their behaviour on the blighted lobes were observed.

After feeding periods of 24 hours, the insects were inactivated by putting them in a deep freezer for 2 hours. After this period, the insects were removed and placed into separate sterile beakers into which the feaces of each insect species were collected (Figure 5). The insects were inactivated in order to allow ease of handling. Approximately 0.5 gm of feaces of each insect species were transferred to sterile test tubes containing 5 ml of sterile distilled water, shaken vigorously and allowed to stand for 30 minutes. In the case of sucking insects whose feaces were always watery, 5 mls of sterile distilled water was pipetted into the beaker, shaken and allowed to stand for 30 minutes before the suspension was poured into the test tube. There was one test tube for feaces of each insect species. After 30 minutes, 1ml of the suspension of feaces of each insect species, was

Fig. 4 : The system used for feeding insects with bacterial blight infected leaves

Fig. 5 : The System used for collecting feaces from insects.

serially diluted three times and 0.1 ml of the final dilution was treated as described above. The remaining suspension was inflitrated into leaf lobes as previously described for A group insects.

3.4 Statistical Procedures

All the data were transformed into square root (X+1) before statistical analysis. The analysis was based on MSTATC programme and was done on IBM PS/2 Model 30 Computer.

The "Analysis of Variance" was the main tool used during the investigation. The Least Significant Difference (LSD) and the Duncan's New Multiple Range Tests were used in seperation of treatment means. However, the "t" test and the Chi-square test were used to compare means for insect preference for bacterial blight infected leaves and to investigate the influence of cassava varieties and seasons of the year on the preference.

Calculations of correlation coefficients were done to investigate the relationship between insects, their feeding sites and bacterial blight lesions, and between bacterial blight lesions and yield and yield components of cassava.

For the first experiment, "two factor randomised complete block design with split-plot combined over locations" module was used in the analysis. Sampling date, varieties and time of day represented locations, cassava varieties and time of the day respectively. The analysis was done on means of each insect species separately. Similarly, the "t" tests were done on each insect species seperately but were based on the combined means of six seasons.

The analysis of variance for the second and third experiment were also based on "Two factor randomised complete block designs with split plot combined over locations" (second experiment only). The sampling dates and the treatments were in the main and sub-plots respectively. For the third experiment, the type of isolate (body surface or feaces) were in the main plots

while isolates from each insect species were in the subplots. A similar arrangement was also done for the infectivity tests for the isolates.

Unless otherwise stated, the asterisks were used to indicate level of statistical significance thus *, ** and *** indicate significance at 0.05, 0.01 and 0.001 levels respectively.

CHAPTER IV

RESULTS

4.1 Insects Associated with CBB

A total of twenty insect species from seven orders were found associated with cassava leaves infected by bacterial blight. The insects species and their numbers on blighted and healthy cassava leaves are shown in Table 3. Appendix 1 gives a pictorial presentation of the species; their taxonomic details appear in Appendix 2.

Total number of insects on cassava leaves infected by bacterial blight were significantly higher than those on healthy ones (Table 3). They remained so at every sampling date throughout the experimental period (Figure 6). Neither varieties nor period of the year affected the distribution of the insects on the cassava leaves (Figures 6 and 7).

Of the insects found associated with cassava leaves infected by bacterial blight (Table 3) Zaprionus sp and Alleculidae were significantly more abundant. And,

Table 3 Insects species and Numbers on Bacterial Blight Infected and on Healthy Cassava Leaves

Insect species	Insects on Healthy leaves	Insects on Diseased leaves
Zaprionus sp	2.66±0.10	10.06±0.25
Alleculidae	1.59±0.09	5.84±0.21
Brunsia occidentalis	1.59±0.11	5.34±0.22
Catantops momboensis centralis	1.59±0.08	4.69±0.16
Paederus sabaeus	1.54±0.14	5.12±0.18
Nematocerus castaneipennis	1.52±0.08	4.71±0.17
Scymus morelleti	1.45±0.09	5.19±0.17
Dermapteran	1.36±0.08	4.63±0.17
Afroeurydemus nigricornis	1.33±0.07	5.16±0.19
Aspavia albidomaculata	1.30±0.07	5.00±0.18
Halydicoris sp	1.30±0.07	5.05±0.19
Chloropidae	1.22±0.07	4.65±0.18
Lagria villosa	1.16±0.07	4.32±0.17
Calidea dregii	1.16±0.07	4.61±0.17
Cautires usambarae	1.15±0.06	4.90±0.18
Creontoides pallidus	1.10±0.07	4.75±0.20
Cheilomenes propinqua vicinia	1.09±0.06	4.67±0.19
Prodagrice sjostedti	1.06±0.06	4.80±0.18
Nezara viridula	1.04±0.06	4.51±0.19
Symploce sp	0.73±0.07	3.73±0.22

Data is based on mean of six recordings.

Symploce sp infested the blighted leaves in significantly lower numbers.

Numerical means for the entire experimental period showed that the times of day: 6.30-7.30 a.m., 12.30-1.30 p.m., or 5.30-6.30 p.m., significantly affected the

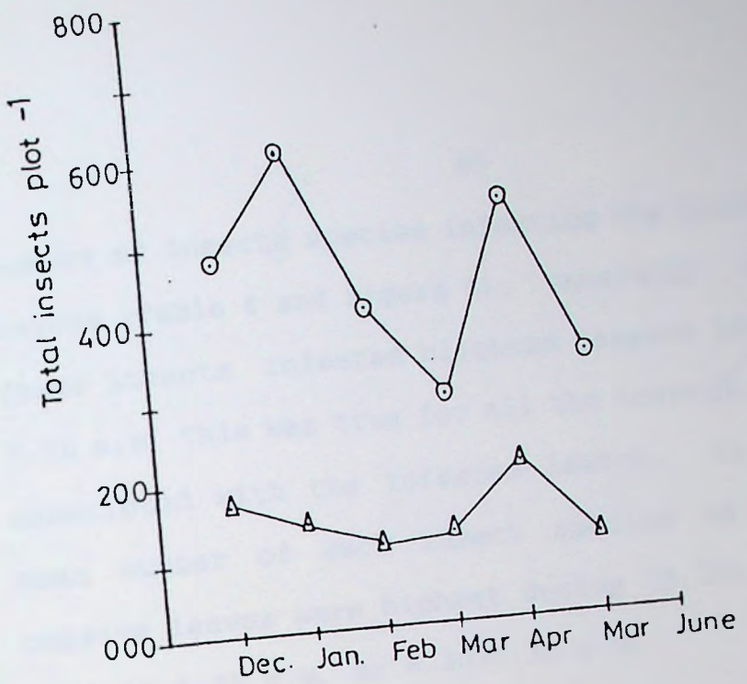

Fig. 6 : Total number of insects on △—△ healthy and on ⊙—⊙ bacterial blight infected cassava at different dates of 1985/86

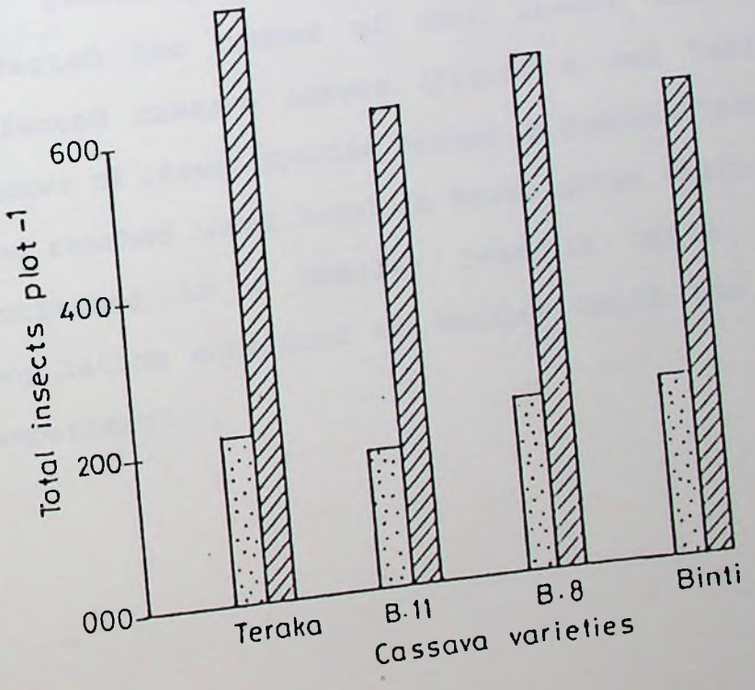

Fig 7 : Total number of insects on ⊡ healthy and on ▨ bacterial blight infected cassava leaves on four cassava varieties: Terka = Ebwanateraka; B8 = Bukalasa 8; B B.11 = Bukalasa 11; Binti = Bintiminti.

number of insects species infesting the blighted cassava leaves (Table 4 and Figure 8). Generally, significantly fewer insects infested blighted cassava leaves at 6.30-7.30 a.m. This was true for all the insects species found associated with the infested leaves. In general, the mean number of each insect species on the infected cassava leaves were highest during 12.30-1.30 p.m. than at 5.30-6.30 p.m. or 6.30-7.30 a.m.

Similarly time of the year also significantly affected the number of each insect species on the infected cassava leaves (Figure 8 and Table 5). The number of insect species peaked in January 1986, declined and reached lower level in March after which they again increased to a smaller peak in April. Thereafter population continued to decline until the end of the experiment.

Table 4 Insect species and numbers on Bacterial Blight Infected Cassava Leaves at three different times of day.

Insect species	Insects at Time of Day		
	6.30 - 7.30 a.m	12.30 - 1.30 p.m	5.30- 6.30p.m
Zaprionus sp	7.04±0.67c	12.25±0.67a	10.89±0.67b
Aspavia albidomaculata	3.62±0.47c	6.17±0.47a	5.20±0.47b
Alleculidae	3.51±0.57b	7.27±0.57a	6.75±0.57a
Scymnus morelleti	3.32±0.43b	6.19±0.43a	6.07±0.43a
Brunsia occidentalis	3.32±0.57c	7.23±0.57a	5.47±0.57b
Calidea dregii	3.09±0.45c	5.64±0.45a	5.10±0.45b
Paederus sabaeus	2.96±0.45c	6.82±0.45a	5.57±0.45b
Cheilomenes propinqua vicinia	2.90±0.49c	6.79±0.49a	4.32±0.49b
Nezara viridula	2.85±0.49c	5.71±0.49a	4.96±0.49b
Afroeurydemus nigricornis	2.82±0.49c	7.10±0.49a	5.57±0.49b
Cautires usambarae	2.76±0.49b	6.27±0.49a	5.67±0.49a
Dermapteran	2.67±0.47b	5.85±0.47a	5.37±0.47a
Symploce sp	2.66±0.51b	4.55±0.51a	4.00±0.51a
Crecontiades pallidus	2.62±0.47c	5.93±0.47a	5.70±0.47b
Lagria villosa	2.62±0.43b	5.11±0.43a	5.22±0.43a
Chloropidae	2.58±0.45b	5.81±0.45a	5.55±0.45a
Halydicoris sp	2.58±0.51c	6.73±0.51a	5.84±0.51b
Podagrice siostedti	2.49±0.47c	7.47±0.47a	4.44±0.47b
Cantantops momboensis centralis	2.30±0.39c	6.32±0.39	5.44±0.39b
Nematocerus castaneipennis	1.91±0.41c	6.64±0.41a	5.60±0.41b

Figures with different subscript within a row are significantly different at the 1% level according toNew Duncan' Multiple Range Test (NDMRT). Data is based on means of six recordings each on four varieties.

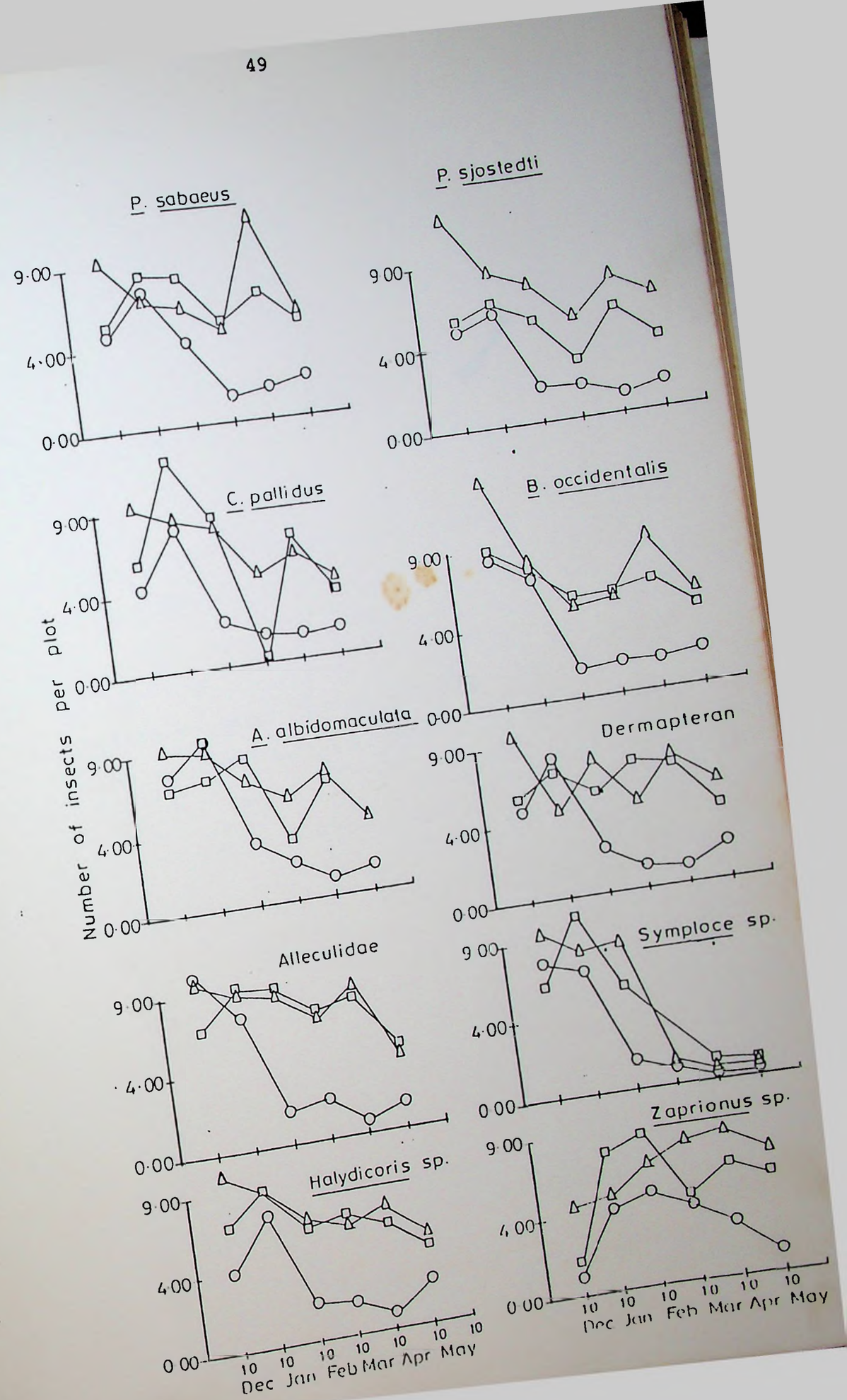

Fig. 8 : The mean number of each of twenty insect species on bacterial blight infected leaves at ○—○ 6.30 - 7.30 a.m, △—△ 12.30 - 1.30 p.m and at □—□ 5.30 - 6.30 p.m of selected months of year 1985 and 1986.

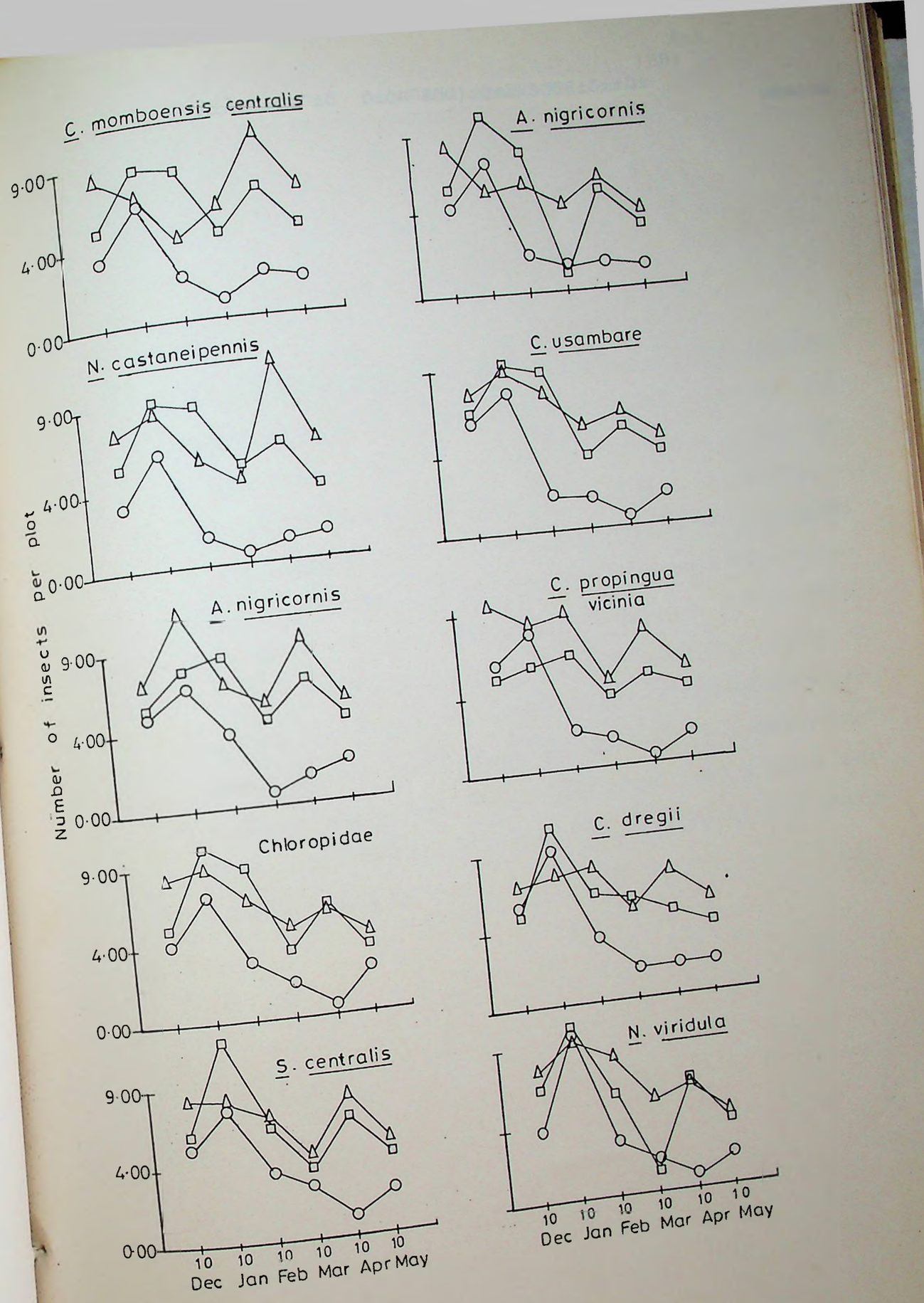

Fig. 8 : The mean number of each of twenty insect species on bacterial blight infected leaves at O—O 6.30 - 7.30 a.m, Δ—Δ 12.30 - 1.30 p.m and at □—□ 5.30 - 6.30 p.m of selected months of year 1985 and 1986.

The relative abundance of each insect species on cassava leaves infected by bacterial blight varied with the time of the year (Table 5). Broadly, species in numerical superiority fluctuated throughout the year. In December, B. occidentalis, Alleculidae. Symplocea sp , P. sjostedti and A. albidomaculata showed similar population trend. Records for January indicated that all but three (Table 5) had built identical population levels. Nearly a uniform decline in population abundance was witnessed in subsequent months although Zaprionus sp still maintained a higher population level. Similarly, marked departure from this trend was observed in Symplocae sp from March to May (Table 5).

Table 5 : Mean Monthly Numbers of Insects on Bacterial Blight Infected Cassava Leaves

Insects species	Dates of Year						
	10/12/85	10/1/86	10/2/86	10/3/86	10/4/86	10/5/86	CV (%)
Catantops momboensis centralis	5.00c	6.44a	4.28d	3.00f	5.79b	3.62e	24.66
Nematocerus castaneipennis	4.65c	7.39a	4.69c	2.71e	5.84b	3.00d	25.95
Paederus sabaeus	5.89c	7.00a	5.53b	2.98c	6.10b	3.12c	26.81
Afroeurydemus nigricornis	5.37bc	8.60a	5.48b	2.98d	5.18c	3.01d	27.63
Scymnus morelleti	5.73b	9.23a	5.22c	3.16e	4.65d	3.19e	25.19
Chloropidae	5.30b	8.53a	5.74b	2.84d	3.22cd	2.25f	28.61
Lagria villosa	5.46b	7.55a	4.88b	2.00e	3.75c	2.27d	28.71
Creontiades pallidus	5.83b	9.58a	5.44c	1.65f	3.80d	2.21e	27.54
Apavia albidomaculata	7.46a	8.12a	5.44b	2.85d	3.95c	2.17d	27.99
Alleculidea	9.14a	8.16b	5.80c	4.46d	5.06d	2.44e	29.87
Cautires usambarae	6.35b	8.70a	5.58c	2.89d	3.32d	2.54e	28.22
Nezara viridula	5.27b	9.37a	5.00b	1.99d	3.26c	2.18d	30.57
Symploce sp	7.79b	8.35a	4.76c	0.66d	0.38de	0.47e	34.17
Cheilomenes propinqua vicinia	6.73a	7.12a	5.33b	2.59d	3.49c	2.76d	30.27
Podagrice sjostedti	7.71a	6.80b	4.58c	2.59e	4.08c	3.02d	29.71
Dermapteran	6.26a	6.10a	4.64c	3.66d	4.19c	2.95d	29.77
Zaprionus sp	6.52c	11.19c	12.19a	10.43c	10.95bc	9.08d	21.65
Brunsia occidentalis	10.68a	7.05b	3.37d	3.38d	4.77c	2.82d	28.60
Halydicoris sp	6.85b	8.04b	4.57c	4.09c	3.75c	3.00d	28.82
Calidea dregii	5.11b	8.53a	4.96b	3.17cd	3.28c	2.62d	27.52

Different subscriptions within a row is significant at 1% (New Duncan's Multiple Range Test)

4.1.3 Insect Behavioural Activities on Diseased Plants

Insect behavioural activities on cassava plants infected by bacterial blight were monitored (Table 6). All the insect species studied seemed to have directional movements towards the infected cassava leaves. Once an insect landed on a plant with the infected leaves, it moved up and down until it found the infected leaves. It then moved to the lesions and preferred to settle on the lower surface of the lesion. This was particularly so with the insects in the order Diptera although insects belonging to other orders could also be found on the lower surface. Once the insect was settled, it fed on the lesion. Insects such as Chloropidae (Diptera) and Zaprionus sp (Drosophilidae) etc. licked and swallowed bacterial exudates or water droplets from the lesions. Lagria villosa (Fab.); Afroeurydemus nigricornis (Selman), Nematocerus castenipennis (Hustache) S. morelleti, Alleculidae; Paederus sabaeus (Erichson), Cautires usambarae (Bourg.); Cheilomenes propinqua vicinia (Mulsant); Scymnus morelleti (Mulsant); Podagrice sjostedti (Jacoby), a Dermapteran, Catantops momboensis

Table 6. Insect behaviour on bacterial blight infected cassava leaves

Insect species	Mean Number of Insects		
	mobile	Feeding	Resting
Zaprionus sp	3.42±0.13	3.48±0.11	3.15±0.07
Alleculidae	2.50±0.13	1.55±0.08	1.81±0.09
Brunsia occidentalis	2.42±0.12	1.38±0.09	1.76±0.10
Scymnus morelleti	2.24±0.11	1.38±0.08	1.57±0.07
Afroeurydemus nigricornis	2.15±0.11	1.40±0.08	1.61±0.09
Cautires usambarae	2.15±0.11	1.35±0.08	1.38±0.07
Halydicoris sp	2.13±0.11	1.36±0.08	1.53±0.08
Paederus sabaeus	2.11±0.11	1.32±0.07	1.68±0.08
Podagrice siostedti	2.09±0.11	1.35±0.08	1.36±0.07
Aspavia albidomaculata	2.07±0.11	1.33±0.08	1.61±0.09
Chloropidae	2.04±0.11	1.22±0.08	1.38±0.08
Cheilomenes propinqua vicinia	2.01±0.11	1.20±0.07	1.48±0.09
Lagria villosa	1.95±0.11	1.07±0.07	1.31±0.07
Creontiades pallidus	1.94±0.11	1.35±0.09	1.48±0.09
Catantops momboensis centralis	1.91±0.10	1.32±0.07	1.47±0.07
Dermapteran	1.89±0.11	1.34±0.08	1.43±0.07
Nematocerus castaneipennis	1.87±0.10	1.30±0.07	1.54±0.08
Calidea dregii	1.84±0.11	1.26±0.08	1.54±0.08
Nezara viridula	1.82±0.10	1.31±0.09	1.37±0.08
Symploce sp	1.65±0.12	1.05±0.09	1.07±0.09

centralis (Jago) Symploce sp and Eulioptera reticulata chewed parts of lesions thus creating "holes" through such lesions.

The feeding behaviour and sites of chewing insects were of two types. For instance, E. reticulata, C. momboensis centralis and L. villosa preferred feeding on edges of lesions or on margins of wilted leaf lobes. They created irregular feeding sites or "holes". Systates castaenipennis preferred feeding on centre of blight lesions or wilted but folded leaf lobes through which they created smooth and round holes. Similarly, Alleculidae, and Afroeurydemus nigricornis, fed only on folded, wilted or blighted leaf lobes thus creating perforations on the lobes.

After feeding or when the weather was unfavourable, the insects either rested on the lower surface of the lesion or walked away and rested on the lower surface of other leaves. They also searched, entered and rested inside wilted and folded leaves or leaf lobes.

4.2 Possible Dissemination of Cassava Bacterial Blight by Insects

4.2.1 Bacterial blight lesions, insect population and insect feeding sites

The effect of molasses, brewers' yeast and dimethoate, on insect population, number of insect feeding sites and bacterial blight lesions were significantly different (Tables 7 - 9).

Total insect population, number of feeding sites and bacterial blight lesions were lowest in dimethoate (Rogor) treated plots than in control plots. Plots treated with molasses and brewers' yeast had the highest number of insects, feeding sites and bacterial blight lesions. However differences betweeen molasses and brewers' yeast treated and those between dimethoate treated and untreated plots were small and insignificant (Tables 7 - 9).

Table 7 : Mean Insect Population visiting cassava leaves after the application of (Dimethoate) and mollasses and brewers' yeast on cassava different times of 1986/87

Treatments	Dates of 1986/87					
	22.11.86	01.12.86	12.12.86	22.12.86	02.01.87	14.01.87
Dimethoate (control)	17.87±2.81a	68.44±3.48a	89.40±2.67a	110.30±1.80a	88.43±1.36a	68.64±2.87a
Unsprayed	24.06±2.81b	87.26±3.48b	115.60±2.67b	103.70±1.80b	87.16±1.36a	69.03±2.87a
Mollasses	24.57±2.81b	159.40±3.48c	170.10±2.67c	186.50±1.80c	176.60±1.36b	147.10±2.87c
Brewers yeast	21.65±2.81b	156.50±3.48c	160.90±2.67d	178.70±1.80d	174.80±1.36b	147.40±2.87b

Figures with different subscripts within a column or row are significant at 5% level of probability according to Duncan's Multiple Range Test (DMRT).

Table 8 : Insect Feeding Sites on Bacterial Blight Lesions After the Application of Dimethoate and Mollasses and Brewers Yeast on Cassava at Different Times of 1986/87

Treatments	Dates of Year 1986/87						
	25.11.86	01.12.86	12.12.86	22.12.86	02.01.87	14.01.87	Mean
Dimethoate (control)	36.46±3.00a	38.72±3.42a	42.36±2.46a	44.68±2.38a	37.06±2.36a	20.45±2.17$_a$	36.62±2.63a
Unsprayed	52.92±3.00b	53.56±3.42b	52.85±2.46b	50.76±2.38a	44.56±2.36a	25.40±2.17$_a$	46.69±2.63b
Mollasses	57.42±3.00b	54.53±3.42b	70.53±2.46c	71.60±2.38b	66.84±2.36b	25.94±2.17$_b$	57.81±2.63c
Brewers yeast	51.41±3.00b	52.48±3.42b	68.49±2.46c	67.44±2.38b	60.69±2.36b	27.42±2.17$_b$	54.66±2.63d

Figures with different subscripts within a column or row are significant at 1% level of probability according to Duncan's Multiple Range Test (DMRT).

Table 9 : Mean Bacterial Blight Lesions on Cassava Leaves after Application of Dimethoate and Mollasses and Brewers Yeast on Cassava at Different Times of 1986/87

	Dates of 1986/87						
Treatments	25.11.86	01.12.86	12.12.86	22.12.86	02.01.87	14.01.87	Mean
Dimethoate (control)	54.74±1.52a	56.11±1.81a	45.82±3.58a	46.53±2.33a	44.01±5.17a	34.44±1.08a	46.94±2.58a
Unsprayed	70.76±1.52b	72.59±1.81b	46.55±3.58a	54.66±2.33b	51.64±5.17a	33.74±1.08a	54.99±2.58b
Mollasses	73.94±1.52b	75.29±1.81b	60.72±3.58b	62.14±2.33c	47.91±5.17a	35.71±1.08a	59.29±2.58c
Brewers Yeast	70.08±1.52b	78.24±1.81b	58.90±3.58b	59.52±2.33b	51.65±5.17a	34.16±1.08a	58.76±2.58c

Figures with different subscripts within a column or row are significantly different at 1% level of probability according to Duncan's Multiple Range Test (DMRT).

Correlations between cassava bacterial blight lesions and number of insects on blighted leaves (r = 0.705**), between bacterial blight lesions and insect feeding sites (r = 0.858**), or between insect feeding sites and the total insect population (r = .905**) were highly significant.

Total insect population, insect feeding sites and bacterial blight lesions all showed highly significant quadratic increase with time although the linear effects were more pronounced in the increases of bacterial blight lesions (Tables 7 - 9). For all treatments except unsprayed control, total insect populations, and the insect feeding sites increased from their lowest levels in late November 1986 and peaked in late December. Steady decline in insect population and insect feeding sites were observed after late December 1986. The lowest numbers were reached at the end of the experiment. The response of the two parameters from unsprayed control were similar but differed by reaching peak levels in mid-December and in early December 1986 respectively.

At the beginning of the onset of treatment (late November 1986) the number of cassava bacterial blight lesions were already high. The number of lesions increased and peaked in early December 1986. They then declined to a lower level in mid December 1986 and rose to a smaller peak in late December 1986. After this period, the number of lesions declined to the lowest levels until the end of the experiment. Generally the increase in bacterial blight lesions were similar to those of insects and insect feeding sites but differed by having two peak periods - a higher one in early December and a smaller one in late December (Table 9).

Mean percentage increase of bacterial blight for unsprayed control, mollasses and brewers' yeast treatments were 33.43%, 56.64% and 41.19% respectively (Apppendix 3). Generally, the changes in percentage increase of bacterial blight with time tended to follow a quadratic trend but the linear effects were more marked. The percentage increase in the number of bacterial blight lesions for brewers' yeast treatments increased from 64.17% in late November 1986 to 96.0% in early December 1986. It then declined until mid

December 1986. A rise to a smaller peak was reached in late December 1986. Later it declined and reached lowest levels on 14 January 1986. For mollasses treatments, the percentage increase in the number of bacterial blight steadly declined until mid-December 1986 after which it rose to a peak in late December and later declined and reached lowest levels in mid-January 1989. Those for unsprayed control followed a similar trend but at all times the percentage increases were lower than for either mollasses or brewers' yeast.

4.3. Potency of Bacterial Isolates from Insects

4.3.1 Abundance of CBB Isolates from Insects

Table 10 shows colonies of X. campestris pv manihotis isolated from body surfaces and faeces of insects associated with cassava bacterial blight lesions.

Table 10. Mean colonies ($x10^3$) of X campestris pv. manihotis isolated from body surfaces and feaces of insects associated with CBB lesions.

Sources	Colonies of X. campestris pv manihotis		
Sterile distilled water (control)	0.00±0.00	-	-
CBB leaf isolate (standard)	8.82±0.06	-	-
Insects	Body surfaces	Feaces	Mean
A. nigricornis	2.01±0.06	4.09±0.06	0.05±0.08ij
Dermapteran	1.02±0.06	4.18±0.06	2.60±0.08fghij
C. momboensis centralis	0.79±0.06	6.25±0.06	3.52±0.08ij
L. villosa	0.62±0.06	3.34±0.06	1.98±0.08cdfghij
N. castaneinennis	0.46±0.06	4.12±0.06	2.29±0.08cdfg
Zaprionus sp	0.29±0.06	3.35±0.06	1.81±0.08cdgh
P. sjostedii	0.13±0.06	3.70±0.06	1.92±0.08bcd
Halydicoris sp	0.08±0.06	0.47±0.06	0.27±0.08ab
S. morelleti	0.00±0.06	3.17±0.06	1.58±0.08cdf
Chloropidae	0.00±0.06	4.36±0.06	2.18±0.08cdfghi
A. albidomaculata	0.00±0.06	2.64±0.06	1.32±0.08ac
C. usambarae	0.00±0.00	2.48±0.06	1.24±0.08cdfg

Figures within columns followed by different subscripts are significantly different at $p < 0.01$ according to Duncan's Multiple Range Test (DMRT).

X. campestris pv manihotis were isolated from feaces of all the insects tested. But isolates from the body surfaces were obtained from 8 species only (Table 10).

The population of the pathogen isolated from the feaces and body surfaces (exoskeletons) of the insects

were significantly lower than those from bacterial blight infected lesions. Although statistically the differences between the sources of bacterial isolates were insignificant, the sample number (2 replicates) was unreliably small. There is however, a clear indication that the populations of the pathogens in the feaces were higher than those on the body surfaces of the insects.

Of the insects studied, A. nigricornis and Dermapteran carried a significantly higher X. campestris pv. manihotis population on their body surfaces. Similarly X. campestris pv. manihotis isolates from feaces of C. momboensis centralis were significantly higher than those from the feaces of any other insect. There were no significant differences between the number of X. campestris pv. manihotis colonies from body surfaces of C momboensis centralis, Dermapteran, N. castaneipennis and L. villosa; or between those from feaces of A. nigricornis, Dermapteran, N. castaneipennis, Chloropidae, L. villosa, Cautries usambarae, S. morelleti, Zaprionus sp and A. albidomaculata.

4.3.2 Infectivity of Isolates

Isolates of X. campestris pv manihotis from feaces of chloropidae, Zaprionus sp, S. morelleti P. sjostedii, C. usambare and from both feaces and body surfaces (exsoskeletons) of Dermapteran, A. nigricornis, A. albidomaculata, N. castaneipennis, C. momboensis centralis and L. villosa produced typical (Lozano and Sequeria 1974; Maraite and Meyer, 1975) bacterial blight symptoms on cassava (Figure 9). The symptoms started as pinpoint watersoaked angular areas on the lower surface of the leaf. These watersoaked points gradually expanded with time, and became visible on the upper leaf surface. The spots enlarged into blight and covered larger areas of the leaf lamina. The affected leaves gradually yellowed, dried and dropped off from the plant, 40±2.5 days after inoculation.

Fig. 9 : Symptoms of cassava bacterial Blight after inoculation of cassava leaves by isolates from either bodysurfaces (exoskeleton) or feaces of insects. The control (a) shows needle pricks at points of inoculation. Fig. (b) shows leaf spots expanding into blights.

The periods (14.5 days) taken by isolates, from feaces and body surfaces of insects, to induce bacterial blight symptoms were much longer than those (5.5 days) by isolates from bacterial blight lesions (Table 11). These periods also differed very significantly ($p = 0.01$) among insects isolates and between isolates from parts of an insect. X. campestris pv. manihots isolates from Lagria villosa, C. momboensis centralis and N. castaneipennis took significantly longer periods (16.25 days, 15.00 days, 14.75 days respectively) to develop bacterial blight symptoms than did those from any other insect. These periods were also significantly different with respect to isolates from body surfaces and from feaces of the same insects. Those from body surfaces took longer time (22.5, 18.5, 18.5 days) than those from feaces (10.50, 11.50, 11.0 days) of the insects. Similarly isolates from feaces of S. morelleti, A. albidomaculata, P. siostedii followed by C. usambarae and C. momboensis centralis took significantly more days to develop bacterial blight symptoms compared to those from feaces of any of the other insects.

Fig. 10a : Scalding of cassava leaves after inoculation of leaves by isolates from body surface of a Dermapteran, (Forticulidae) five days after inoculation.

Fig. 10b : Scalding and tattering of cassava leaves after inoculation of the leaves by isolates from body surface of Dermapteran (Forticulidae) - days after inoculation.

CHAPTER V

DISCUSSION

5.1 Insects Associated with CBB

5.1.1 Insect preference for cassava leaves infected by bacterial blight

Insects associated with cassava plants infected with bacterial blight are diverse. In this study, nine Colepterans, five Heteropterans, two Dipterans, two Orthopterans and one each of Hymenopteran, Dictyopteran and Dermapteran Dipteran species were recorded on four cassava varieties screened for susceptibilities to CBB. Such a diverse group of insects suggest that most insects, particultarly those in the orders recorded, may infest cassava plants infected by bacterial blight.

The significantly high number of insects on the infected cassava leaves throughout the period of the experiment strongly suggests insect-bacterial blight association. It also suggests that insects preferred the infected leaves to healthy ones. And evidence indicate that this preference is neither influenced by

host varieties nor seasons of the year. Terry (1974, 1978), Lozano (1975) and Nyiira and Otim-Nape (1978) also found increased insect infestation of cassava leaves and plants infected by bacterial blight. Lesions of bacterial blight Xanthomonas campestris pv. phaseoli, X. campestris pv. phaseoli var fuscans, X. campestris pv. phaseoli f.sp vignicola of beans (Kaiser and Vakili, 1978) and collapsed inflorescence, fruits, heart rots of pineapples, Erwinia chrysanthemi, (Lim and Lowlings, 1977) elicited similar response. The reasons for the insects preference for bacterial blight infected cassava leaves is still unclear although Hasanali (1978) in personal communicatin suggested that the insects may be attracted to the infected leaves and lesions by kariomones emitted from the lesions.

The sheer number and diversity of the insects associated with cassava leaves infected with bacterial blight suggests that the attractant is not specific. The search for this attractant must therefore be diverted towards a non-specific resource which may not necessarily be a kariomone in nature although Hasanali (date) thinks otherwise.

Associations between coleopterans (Stewart 1913a; Rand 1923; Carter, 1973; Kleopper, 1979 and Daniel et al., 1981), Dipterans (Kleopper, 1979; Lim and Lowlings, 1973), Orthopterans (List and Krutzner, 1942 and Daniel et al., 1981) and various bacterial diseases of plants have been reported. Nevertheless, association between bacterial plant diseases and Heteropterans, Hymenopterans, Dictyopterans and Dermapteran insect species was not discernable (Kaiser and Vakili 1978; Buddenhagen and Elsassers, 1962 and Buddenhagen and Kalman 1964 and Revilla, 1967).

We cannot for sure generalize that all the twenty odd insect species found associated with diseased cassava host disseminate CBB. For example, Zonocerus variegatus which has been implicated in the dissemmination of CBB (Daniel et al. ,1981) is indeed a pest of cassava. Thus, the interaction of the two Orthopterans (E. reticulata and C. momboensis centralis) with cassava hosts may be viewed from two stand-points: the visits for CBB lesions as a resource, or to cassava as a host plant. This might as well be true for other insect species whose roles were never explicitly investigated during this study.

Insect species which exploit the two resources concurrently may be of special interest in the epidemiology of CBB. While it is true that the principal way of CBB dissemination is through defication (Tables 10 and 11), bacteria lodging in the buccal cavity of such phytophagous insects may immensely augment infection. For cassava host plants infested with such insects, the physiological impact on the host plant could be more deleterious compared to the species which exploit cassava in only one of the two ways. Consequently, yield differentials may accrue from such simple partitioning of resource exploitation.

The significantly high number of insects on cassava leaves infected by bacterial blight at 12.30-1.30 p.m. is expected. This period is the warmest time of the day in the tropics when temperatures are high. And warm temperature favours insect activity (Dunn and Wright 1955, Cooke 1963, Campbell and Mackauer 1975, Hutchinson and Hogg 1984). In a number of instances, the residual effects of temperature were obviously extended late in the evening (Table 4) especially in the relatively drier months (Figure 8) as discussed below. Early morning

(6.30-7.30 a.m.) low temperature ensures that insect activity is reduced to a minimum. Insect activity is known to be correlated with disease transmission. Kleopper et al (1979) found lowest and highest insect transmission of Erwinia carotovora var carotovora and E. carotovora var atroseptica during the periods 6.00 a.m.-4.00 p.m. and 12.00 -10.00 p.m. respecively.

5.1.2 Temporal Activities of Insects on Blighted Cassava Leaves

The significant effect of time of year on most insect species infesting blighted cassava leaves is suggestive of environmental influences.

Several studies on the dynamics of insect populations have reported peaks depending on seasons (Dun and Wright 1955, Cooke 1963, Smith and Hagen 1966, Clark et al. 1967, Pass and Parr 1971). In the present study two peaks for most of the insect species, - a major one in January and a smaller one in April 1986, were observed. Examination of the weather data (Figure 11) and bacterial blight lesions for the study period

(December-May) revealed that population trends for most of the insect species generally corresponded to the pattern of rainfall. The favourable rainfall during these periods could have stimulated more cassava leaf growths which in turn offered more infection sites for bacterial blights. The increased bacterial blight lesions must have, in turn, attracted more insects and had a positive influence on their survival and reproduction. The higher nutrient value of leaves and lesions (McKee 1962) of plants during favourable rainfall can have a positive influence on survival and reproduction in phytophagous insects (McNeill and Southwood 1978). Culliney et al (1988) recorded highest counts of Pineus pini Macquart and Leucopis obscura Holiday at times of high precipitation, and low number of H. pini and L. obscura were recorded at times of reduced rainfall.

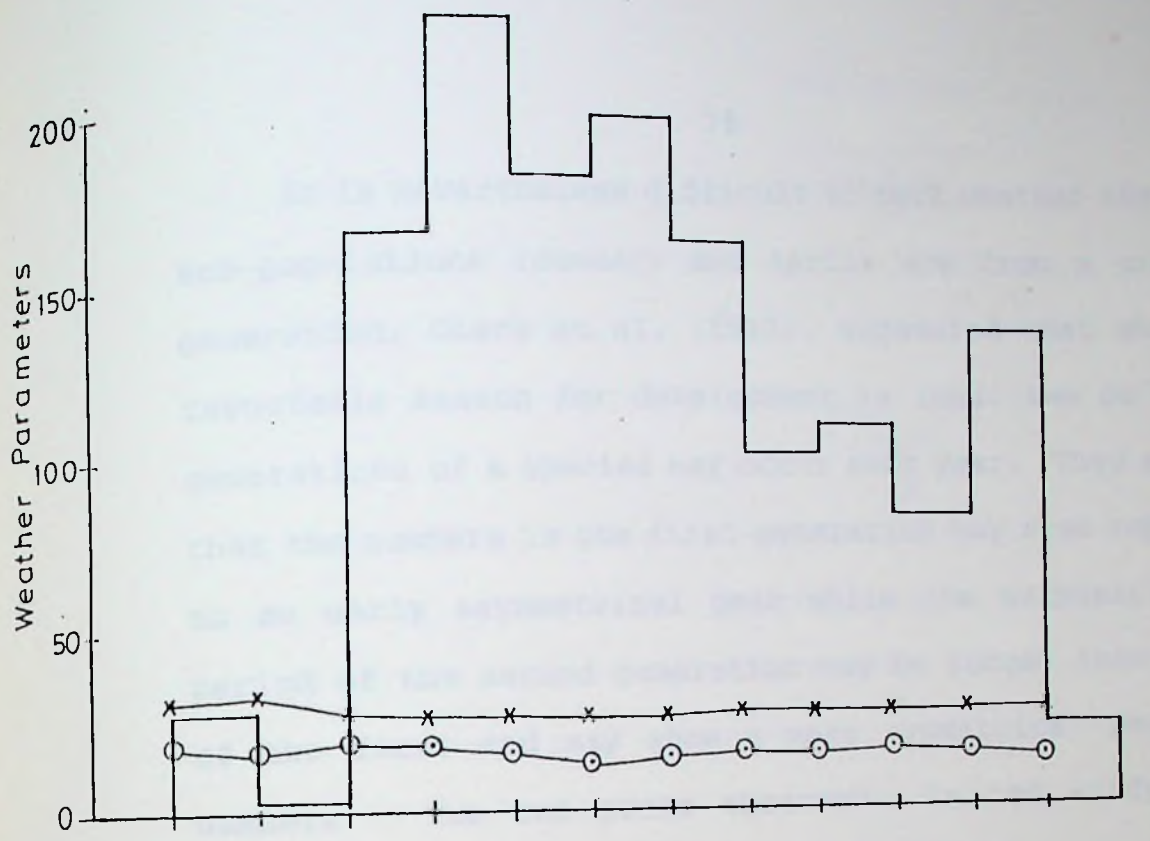

Fig. 11a : Recorded weather data ☐ monthly rainfall (mm) o—o minimum and x—x maximum monthly temperature (^{o}C) during the experimental periods 1985.

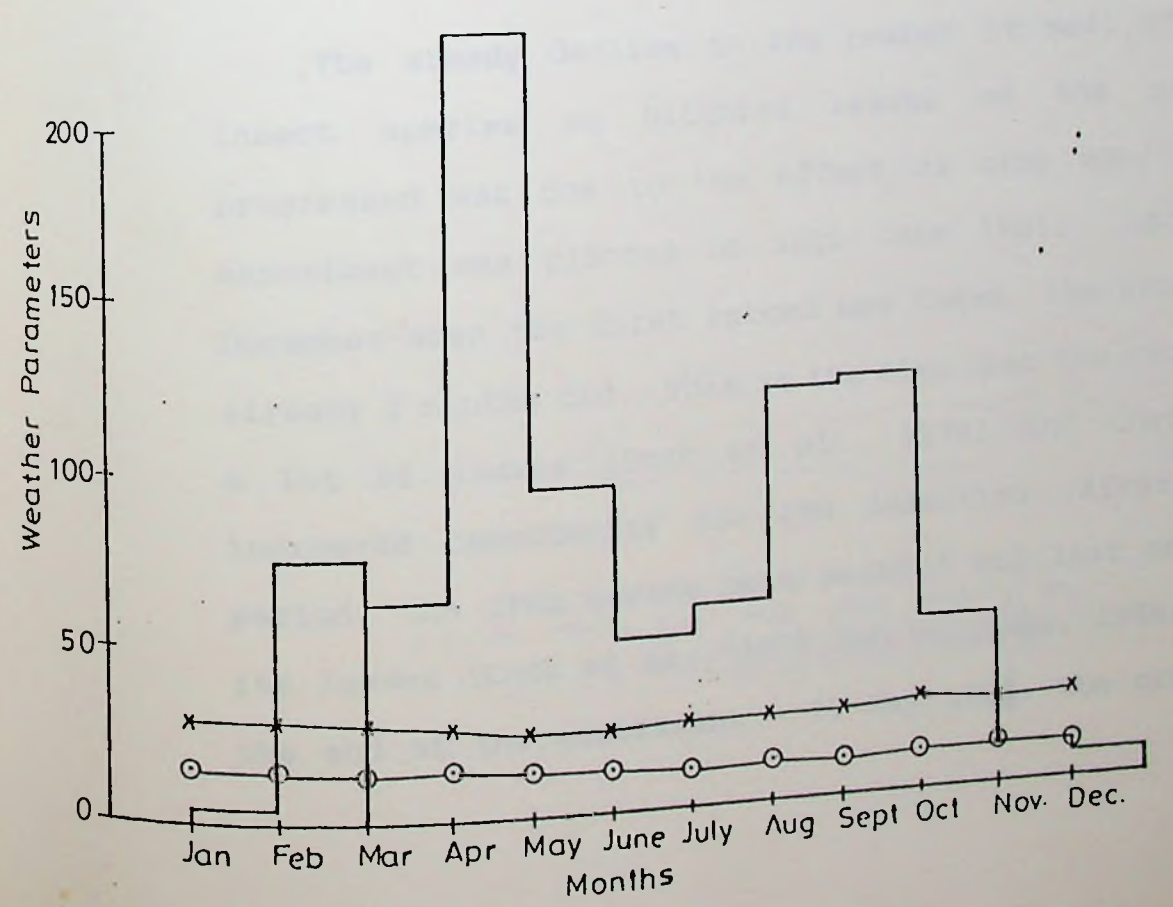

Fig 11b : Recorded weather data ☐ monthly rainfall (mm) o—o minimum and x—x maximum monthly temperatures (^{o}C) during the experimental periods 1986.

It is nevertheless difficult to tell whether the two sub-populations (January and April) are from a single generation. Clark et al. (1967), suggested that when a favourable season for development is long, two or more generations of a species may occur each year. They added that the numbers in the first generation may rise rapidly to an early asymmetrical peak while the ovipositional period of the second generation may be longer than that of the first and may show a more symmetrical peak of numbers. The two peaks observed in the study may suggest different generations of the insects.

The steady decline in the number of most of the insect species on blighted leaves as the season progressed was due to the effect of crop age. The experiment was planted on 10th June 1985. And, by December when the first record was taken, the crop was already 6 months old. This is the time when the crop had a lot of leaves (Cock et al., 1979) and therefore increased opportunity for CBB infection. After this period, the crop became more matured and lost more of its leaves (Cock et al., 1979 and Veltkamp, 1985). By the end of the experiment - 10 May 1986, the crop was

twelve months old, had matured and lost a lot of its leaves. The changes in foliage could have affected the number of bacterial blight lesions which in turn affected the number of insects which visited the infected leaves.

Differential responses observed on some species e.g. Alleculidae, Dermapteran, B. occidentalis and Zaprionus sp (Figure 8) is difficult to explain due to the preliminary nature of the data. And yet this departure appears to be an important aspect of the ecology of the insect community on blighted cassava.

5.1.4 Insect Beavioural Activities on Diseased Plants

The association of insects with blighted cassava leaves suggests a possible role of insects in the epidemiology of cassava bacterial blight. The mode of insect transmission of CBB was nevertheless not fully investigated in this study. But in Zaire (Maraite and Meyer 1975) and Congo (Daniel et al., 1981) infection of cassava by bacterial blight always originated from injuries and punctures made principally by Pseudoteraptus devastans and Z. variegatus respectively.

And, in Puerto Rico, bacterial blight of beans and cowpeas frequently were associated with insect feeding injuries (Kaiser and Vakili 1978). It is likely that while feeding on lesions, the body surfaces and alimentary canal and feaces of the insects may become contaminated with the pathogen. The pathogen may then be disseminated as insects move from plant to plant. The strong correlations between the insects and bacterial blight lesions ($r = 0.71^{**}$), and between the insects and feeding sites ($r = 0.9^{**}$) strongly support the above inference and emphasize the possible role of insects in the dissemination of the disease.

The observed directional movement of insects towards lesions appear to be mediated by stimuli eminating from the lesions. And, as alluded to earlier, these stimuli are probably non-specific in nature.

5.2. Possible Dissemination of Bacterial Blight by Insects

5.2.1. The effects of insect attractants and dimethoate on bacterial blight lesions, insect populations and feeding sites.

The significant increase in insect population on cassava treated with mollasses and brewers' yeast clearly indicates that the products attracted insects from inoculum sources (spreader plants infected by bacterial blight) to test plants. This population increase was accompanied by a similar magnitude of increase in bacterial blight lesions, suggesting that the immigrant insects were actually vectors of the disease. Further evidence were borne out by significant correlation between (i) total insect population and insect feeding sites ($r = 0.91^{**}$), (ii) total insect population and bacterial blight lesions ($r = 0.71^{**}$) and (iii) feeding sites and bacterial blight lesions ($r = 0.86^{**}$).

Although the attractancy of CBB is yet uncertain, this set of experiments (mollasses and brewers' yeast) provides an important clue towards the understanding of the ecology of this disease.

Whether the insects initiate bacterial blight or they are attracted by bacterial blight is also unclear. It has been observed that bacterial blight infections always start from injuries made by insects or by mechanical means. Feaces of insects have been observed on cassava leaves. It is likely that initially the insects get attracted to bacterial blight lesions but once a good number of lesions have been generated, the insects continue to visit them and disseminate the pathogen.

Dimethoate reduced but did not completely eliminate the insects visiting the plants. Its use, however, enabled a satisfactory estimation of the role of insects in the dissemination of bacterial blight under natural conditions (Tables 7 - 9). It also indicated the potential for the use of insecticide in minimizing the spread of the disease by insects. Any programme aimed at

reducing insect population on cassava by use of insecticide will result in reduction of the rate of spread of the disease. Nevertheless it should also be realised that the epidemiology of this disease also involved other factors such as wind and rainsplash (Lozano and Sequeria, 1974b) and therefore the disease intensity witnessed in Dimethoate plots could have been due to these factors. Without recourse to insecticidal intervention, it is possible that between 35.6-56.6% of CBB infection may be effected by insects alone (Appendix 3). This estimate nevertheless is in sharp contrast with those of CIAT (1973), Terry (1974) and Lozano (1975) which ranged from 10-14%.

Indeed the ecology of CBB may not be fully appreciated without cognizance of host plant phenology. Thus insect population increase observed in this study may be partly explained by the phenological events occuring during the experiment. A clear linkage between host plant physiology and insect population was evident throughout the study period (Figure 8; Tables 5, 7, 8 and 9).

5.3 Potency of Bacterial Isolates from Insects

The implications of the present study are important in understanding the epidemiology of cassava bacterial blight. This study has undoubtedly demonstrated that some insects carry and disseminate Xanthomonas campestris pv manihotis through their feaces and body surfaces (exoskeletons). The finding agrees with works of Kaiser and Vakili (1978), Graham et al. (1976), Harrison et al. (1977), Phillips (1977) and Tamini and Brandfield (1979).

Although the insects carry and disseminate the pathogen through their feaces and body surfaces, they carry a lower pathogen population than those found in lesions infected with bacterial blight. These differences, particularly those from feaces, may be due to a reduction of the population of the pathogen as a result of adverse environmental conditions. The bacterial inoculum carried on the exoskeleton of naturally infested insects or present in their feaces is probably reduced quickly to low levels through dessication or exposure to ultra-violet radiation (Kaiser and Vakili 1978). Apparently a similar high mortality

also occurs on the cells of other bacterial plant pathogens on the surface of plant tissues (Leben 1963, 1974; Scuster and Coyne 1975).

The results of the study tend to suggest that feeding behaviour seem to determine how and in what quantity an insect may carry and disseminate the pathogen. Leaf-chewing insects apparently carry and disseminate the bacteria both on their body surfaces and feaces while leaf sucking ones tend to do so in their feaces only. This difference may be due to the fact that the insects are not normally pests of cassava but they walk and feed on bacterial blight lesions. It is most likely that during this process, cells of the pathogen lodge on the appendages and mouthparts of the insects and get disseminated as the insects move from one leaf or plant to another. In addition, these insects ingest the bacteria as they feed on bacterial blight lesions. The data suggest that most of the ingested bacteria pass unaffected through the alimentary system of the insect and are excreted in the feaces. It is likely that the feaces further protect the bacteria from dessication and from ultra violet radiation during adverse weather

conditions and ensures their survival until favourable condition for infection. This fact may explain why the population of the pathogen tended to be higher in the feaces than on the body surfaces of the insects.

How the ingested cells of the pathogen pass through the guts of the insect unaffected by the digestive enzymes of the insects need further investigation. A symbiotic relationship similar to that between Pseudomonas sevastanoi - casual agent of Olive knot, and the Olive fruit fly Dacus oleae (Gmelin) (Hagen et al 1963, 1966; Fytizas and Tzanakakis 1966a, b and c; Fytizas 1969a and b and Rey 1969) may be possible.

Compared to other insects, the population of X. campestris pv. manihotis were higher on body surface of A. nigricornis. This difference is probably due to the insect's ecological and feeding behaviour on cassava leaves infected by bacterial blight. A. nigricornis is a small brown and ovalish beetle that likes hiding itself and feeding on folded leaves infected by bacterial blight. It is likely that this constant association with bacterial blight lesions exposes the insect to a lot of

contamination with the pathogen. Hiding in folded infected leaves also protects it from adverse environmental conditions and ensures high survival of the pathogen on its exoskeleton.

Similarly the high population of the pathogen in the feaces of C. momboensis centralis may be due to the feeding habit of the insect. The insect feeds voraciously (unpublished, personal observation) on large areas of bacterial blight lesions and consequently excretes bigger feaces. Its feeding habit enables it to ingest high proportions of the pathogen. The bigger feaces could also offer better protection to the bacterial cells in them. Furthermore, the alimentary system of the insect could have ensured that the bacterial cells of the pathogen passed through it with as little destruction as possible.

Development of symptoms varied with the source of isolate (Tables 10 and 11). Probable differences in pathogenic load of the various isolates may explain this anomaly. This has important bearing on one of the cultural strategies of pest management, namely delayed

planting time (Stern et al, 1976). Under this scenario availability of inoculum source is obviously spread over a much longer time thus increasing the potential for disease acquisition by individual insects that would have otherwise missed the opportunity to do so. This is further exacerbated by the semi-perennial nature of cassava.

CHAPTER VI

SUMMARY AND CONCLUSION

One of the main requirements in the control of plant diseases is an adequate knowledge of the epidemiology of the disease. This necessitates an indepth understanding of each of the factors in the epidemiology. Insects have been implicated in the epidemiology of some bacterial blight disease (Amaral, 1915; Maraite and Meyer, 1975; Nyiira and Otim-Nape, 1978). In this thesis evidence is presented on the role of insects in the epidemiology of CBB.

Twenty insect species from seven orders were found associated with bacterial blight lesions on cassava leaves. Such associations were enhanced by warmer weather. Lim and Lowlings (1977) and Kloepper *et al*. (1979) also observed large species of insects associated with collapsed pineapple fruits and potato blackleg diseases respectively. As already hinted throughout this thesis, such a large group of insects certainly do have important implications in the ecology of CBB. This is to be expected because afterall the success of insects as vectors of plant pathogens depends on insect numbers and

activities (Buddenhagen and Elsessers, 1962). But this must be backed-up by the abundance, viability and virulence of the pathogen and the frequency and susceptibility of tissues for infection (Buddenhagen and Elsessers, 1962). The later aspect of CBB epidemiology was demonstrated by the results of the experiment which showed diffential virulence of CBB isolates: speculated to be due to differences in pathogen load (abundance).

With the exception of C. momboensis centralis, the insects found associating with blight lesions are not pests of cassava (Daniel et al., 1981). Thus at best, their attraction to CBB lesions signify a commensual relationship. Unfortunately, the cassava does not benefit from this relationship due to the fact that in the course of the association the insects disseminate the disease to healthy plants and leaves. The relationship ensures that the insects get in touch with the disease and acquire the pathogen.

The contribution of insects to the epidemiology of CBB as deduced from the data at hand shows unexplainable discrepancy with the earlier estimates (CIAT, 1973;

Terry, 1974 and Lozano, 1975) which suggest insignificant roles of insects in the dissemination of the disease. Nevertheless, the discrepancy must be resolved in a well planned investigation. Indeed the role of insects in the epidemiology of CBB could be profound when we consider the induction of the disease in an isolated and healthy cassava fields. Although, ordinarily, the pathogen could be transmitted by wind, rainsplash, infected materials, true seeds, plant debris and volunteer plants, the scenario given above rules out their roles and leaves insects as the sole vectors for initiating initial infection in such fields. This indeed is a controversial proposition given that most of the insects associating with blighted cassava leaves are actually not pests and therefore can not cause mechanical injuries through which the pathogens can gain entry into the plant and cause infections. We however note that Xanthomonas campestris pv manihotis, the CBB pathogen, can survive as epiphyte on cassava leaves and cause infection during favourable conditions (Daniel and Bohr, 1978; Persley, 1978). Under this condition, the major source of pathogen would come from feaces (Tables 10 and 11).

Consequently it becomes extremely important to reduce potential sources of bacterial blight inoculum (Kleopper, 1981) and the rate of dissemination of the disease. Although the application of insecticide (Dimethoate) did not completely eliminate the insects and bacterial blight infections, its use however reduced the number of insects and the subsequent bacterial blight lesions to low levels. Maintenance of a reasonable insect control programme would therefore help to reduce the level of the disease in the field. An integrated approach combining the use of crop rotation, use of resistant varieties, bacterial blight-free planting stems, destruction of volunteer cassava plants in neighbouring areas, destruction of isolated infected plants within the field, and the implementation of a practical insect control programme could reduce bacterial blight in Uganda to a low level.

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Appendix 1

Insect Codes	Insect Names
1	Creontiodes pallidus
2	Lagria villosa
3	Eulioptera reticulata
4	Alleculidae
5	Aspavia albidomaculata
6	Scymnus morelleti
7	Chloropidae
8	Paederus sabaeus
9	Nematocerus castaneipennis
10	Afroeurydemus nigricornis
11	Catantops momboensis centralis
12	Halydicoris sp
13	Brunsia occidentalis
14	Zaprionus sp
15	Dermapteran
16	Podagrice sjostedti
17	Cheilomenes propingua vicinia
18	Symploce sp
19	Nezara viridula
20	Cautires usambarae
21	Calidea dregii

Appendix 3 : Estimated % of Bacterial Blight Infectation

	Time of Year 1986/87						
Treatments	25.11.86	01.12.86	12.12.86	22.12.86	02.01.87	14.01.87	Mean
Dimethoate (Control)	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Unsprayed	67.50	64.46	25.90	41.70	8.86	4.17	35.43
Mollasses	82.50	80.20	74.10	81.50	21.52	0.00	56.64
Brewers Yeast	64.17	96.00	63.50	27.92	36.71	0.00	41.19