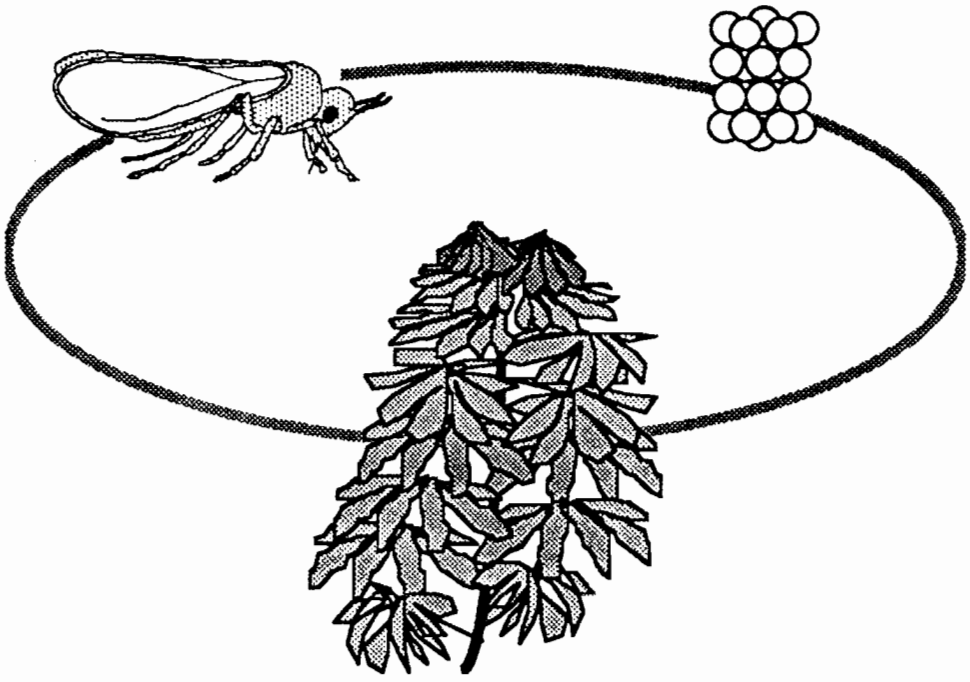
INSTITUT FRANÇAIS DE RECHERCHE SCIENTIFIQUE POUR LE DEVELOPPEMENT EN COOPERATION
BP. V 51 ABIDJAN (COTE D'IVOIRE)

SOME OBSERVATIONS
ON
BEMISIA HANCOCKI (CORBETT)



Research report of
Teus G. van Lingen and Godwin A. Limberg. **May 1988**

under the direction of Dr. L. D. C. Fishpool and Dr. C. Fauquet
Virology department, ORSTOM.

CONTENTS

PREFACE

As part of our MSc studies we undertook a research project on the whitefly *Bemisia hancocki* (Corbett), (Homoptera, Aleyrodidae), a close relative of the important pest *Bemisia tabaci* (Genn.). *B. tabaci*.. is a known vector of many viruses (for example, Okra Leaf Curl Virus, Tomato Yellow Leaf Curl Virus (Sharaf, 1982), and Cotton Leaf Curl Virus (Mound, 1983)). In 1980 ORSTOM (*Institut Français de Recherche Scientifique pour le Developpement en Cooperation*) began an investigative study of African Cassava Mosaic Virus or ACMV, also vectored by *B. tabaci*. This work has been undertaken in Ivory Coast, in an area where cassava is the basic staple food and where considerable yield losses attibutable to ACMV, occur annually (Fauquet & Fargette, 1987). Initially the programme concentrated largely on the virus and the plant itself but in 1986 a more detailed study of the vector was begun. This work is concerned mainly with an investigation of the population dynamics of *B.tabaci*. We participated in the entomological research programme from January to July 1988. We made a study of *B. hancocki*, to try to establish whether this species is also a vector of ACMV.

The structure of this report is as follows: in Chapter One an introduction to the local conditions is given. Also the exact aims of our study are stated. The second chapter is an outline of the literature involving the biology of both *Bemisia* species. In the third chapter the methods used are described and the succeeding chapters are dedicated to the presentation and discussion of the results. A seperate report will deal with the natural enemies of *B. tabaci* and their efficiency in controlling this whitefly.

We would like to express our thanks to all people that made this study possible. We are especially grateful to Dr. L.D.C. Fishpool and to Dr. C Fauquet, our tutors and to Dr. J.-C. Thouvenel, who took care of our needs whenever necessary. Also we are grateful to Dr. W. Takken and Dr. A. van Huis, our supervisors, who made the necessary contacts thus making this research possible.

SUMMARY

This study deals with the problem of the transmission of African Cassava Mosaic Virus by *Bemisia hancocki*. This is a whitefly species (Homoptera, Aleyrodidae), which occurs in relatively low numbers in cassava fields in Ivory Coast. The numerical importance of this species as compared to the congeneric *B. tabaci*, as well as the capacity of *B. hancocki* to transmit the ACMV, is investigated.

To assess the numbers of *B. hancocki* in the field, counts of the nymphs were made in two experimental cassava plots. From these counts it appeared that the species is rare compared with *B. tabaci*. When the crop is young the numbers of *B.hancocki* are about 5% of the total nymphal whitefly population, but increase relatively from around the 120th day after planting. This probably results from a large relative decrease in *B. tabaci* numbers at this time. Mean numbers ranged from 0 to 60 late instar *B. hancocki* nymphs to 190 to 590 late instar *B. tabaci* nymphs per plant. The distribution of the nymphs of both species within the field follows the same pattern, with larger populations being recorded at the upwind field borders.

The establishment of a pure laboratory culture of *B. hancocki* did not succeed, because of heavy contamination by *B. tabaci*. The source of this contamination was not indentified. It seems that the development of *B. hancocki* nymphs is relatively slow and care has to be taken to prevent the potted laboratory plants from shedding their leaves too early.

Unfortunately the transmission test plants dried out before all observations had been made. Only an indication was found that the ACMV can be transmitted by *B. hancocki*. Further trials are necessary.

RÉSUMÉ

Cette étude s'occupe avec la problème de la transmission de la Mosaïque Africaine du Manioc (ACMV) par *Bemisia hancocki*. Il est une espèce de mouche blanche, qui se présente en numéros rélativement bas, comparés avec celles de l'espèce rélaté *Bemisia tabaci* dans les champs du manioc de la Côte d'Ivoire. L'importance de cette espèce comparée à *B. tabaci* est examiné, de même que sa capacité de transmettre le virus.

Pour faire une estimation des numéros de *B. hancocki* dans le champ, comptes ont été fait dans deux champs expérimentals. Ces comptes ont élucidé que cette espèce est rare, comparé à *B. tabaci*. Les numéros moyennes de *B.hancocki* sont bas, comparé à le numéro total de larves de *Bemisia* sp. Quand le champ est jeune, les numéros se trouvent autour de 5% et ils augmentent après le 120ème jour. après la plantation du manioc. Celà n'est pas la résulte d'une augmentation de la fécundité de *B. hancocki*, mais seulement une diminuation plus rapide des numéros de *B. tabaci*. Les numéros moyennes se trouvent entre 0 et 60 larves des stades plus agées vivantes de *B. hancocki* et 190 jusq'a 590 larves des stades plus agées vivantes de *B. tabaci* par plante. La distribution des mouches dans le champ suive la même modèle. *B. hancocki* et *B. tabaci* groupent sur les limites exposées au vent.

L'établissement d'une culture pure de *B. hancocki* n'a pas réussi bien, à cause d'une contamination grave avec *B. tabaci*. La source de cette contamination n'a pas été révélé. Il est probable que la dévéloppement larval de *B. hancocki* est assez lente. Il faut qu'on s'assure bien de bien traiter les plantes pour préventir la perte des feuilles.

Malheureusement les plantes dans l'expériment de transmission du ACMV ont été sèchée avant qu'était possible de faire toutes observations. Il existe seulement l'indication de qu'il est possible pour *B. hancocki* de transmettre le ACMV. Il reste necessaire de refaire cettes expériments.

SAMENVATTING

Dit onderzoek gaat in op het pobleem van de transmissie van het Afrikaans Cassave Mozaïek Virus (ACMV) door *Bemisia hancocki*. Dit is een witte vlieg, die in relatief lage aantallen voorkomt in de cassave velden van Ivoorkust. Het belang van deze soort, vergeleken met de nauw verwante soort *B. tabaci,* evenals het vermogen om ACMV over te dragen werd onderzocht.

Om een schatting te kunnen maken van de aantallen *B. hancocki* nymphen die in het veld aanwezig zijn, werden tellingen verricht aan twee experimentele velden. Uit deze tellingen werd duidelijk dat de soort in vergelijking tot *B. tabaci* weinig voorkomt. De gemiddelde aantallen zijn laag, vergeleken bij het totaal aantal *Bemisia* nymphen. In het jonge veld maakt *B. hancocki* 5% van het totaal uit. Na de 120ste dag stijgt dit percentage, niet zozeer als gevolg van een hogere voortplantingssnelheid van *B. hancocki,* alswel een snellere afname van de populatie *B. tabaci* nymphen. De gemidddelde populatie van laatste stadium nymphen van *B. hancocki* liep van 0 tot 60 levende nymphen per plant en die van *B. tabaci* liep van 190 tot 590 levende nymphen per plant. De verdeling van de populatie over het veld vertoont voor beide soorten hetzelfde patroon. De vliegen klusteren aan de bovenwindse zijde van het veld.

Het opzetten van een reincultuur van *B. hancocki* is niet gelukt, door een zware besmetting met *B. tabaci*. De bron van deze besmetting is niet gevonden. Het schijnt dat de ontwikkelmingstijd van *B. hancocki* larven lang is. Daarom dient er zorg voor gedragen te worden, de planten in de cultuur te behoeden voor vroege bladval.

Ongelukkigerwijs zijn de planten van de transmissietest verdroogd voor alle waarnemingen gedaan konden worden. Er is uitsluitend een indicatie verkregen dat het ACMV door *B. hancocki* overgebracht kan worden. Het blijft noodzakelijk om verdere proeven te ondernemen.

1. INTRODUCTION

In large parts of Africa cassava (*Manihot esculentus)* is an extremely important food crop. One of the problems confronting the cultivation of cassava in Africa is the disease caused by African Cassava Mosaic Virus (ACMV) which is capable of inflicting substantial crop losses. The virus is transmitted by man and through the continued and widespread planting of contaminated cuttings, the virus has become widespread. In addition the virus is transmitted by the whitefly *Bemisia tabaci* (Homoptera; Aleyrodidae) This report is concerned with a second species *Bemisia hancocki,* which may also be a vector of ACMV.

1.1. Description of the environment

The work described in this report was carried out at the ORSTOM field station of Adiopodoumé situated in the forest zone of southern Ivory Coast. The site is located some 20 km west of Abidjan. In this section some climatological data are presented. Only those data that influenced the research are discussed. The most important factor is the rainfall. In Fig. 1.1 the average monthly rainfall is shown. These are averages over the past 40 years. In the same figure the rainfall during the research period is shown. Fig. 1.1 makes clear that the rainfall during the months January to May 1988 was below normal. The influence of the rainfall on whitefly numbers will be discussed in Chapter 3.

Fig. 1.2. shows the average monthly field temperatures: mean, maximum and minimum. The high maximum temperatures posed serious problems to the establishment of a pure culture of *B. hancocki* (see Chapter 4).

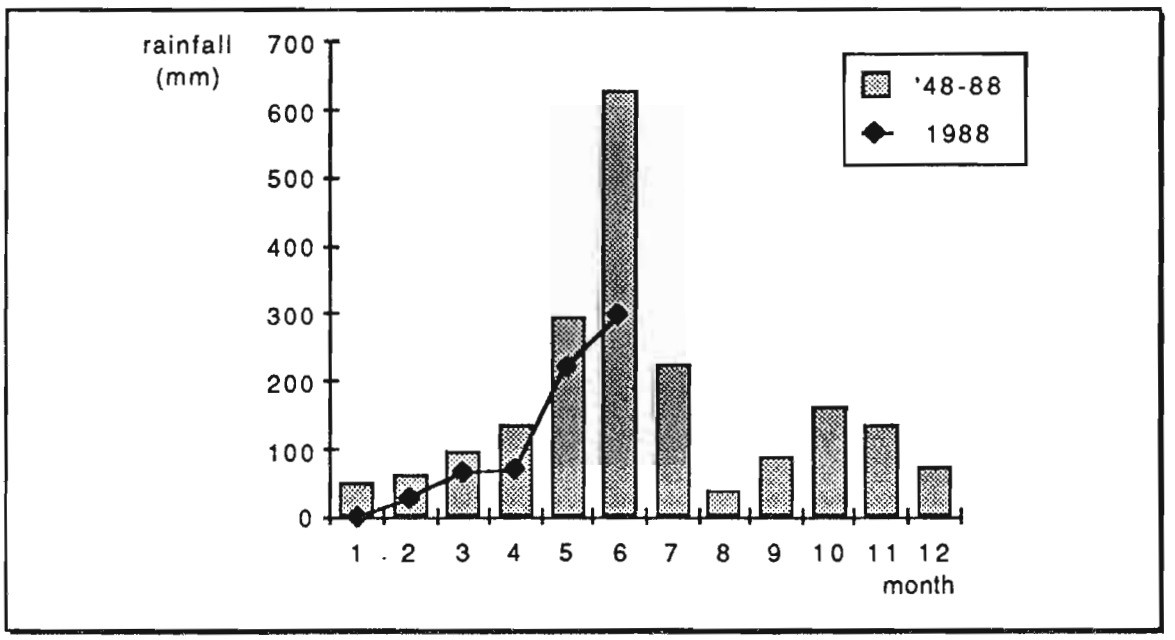


Fig. 1.1. Distribution of rainfall at Adiopodoumé; average monthly rainfall over the past 40 years and rainfall in 1988 (January - June) - *pluviometrie à Adiopodoumé; moyenne de 40 années et pluviometrie en 1988 (janvier - juin).*

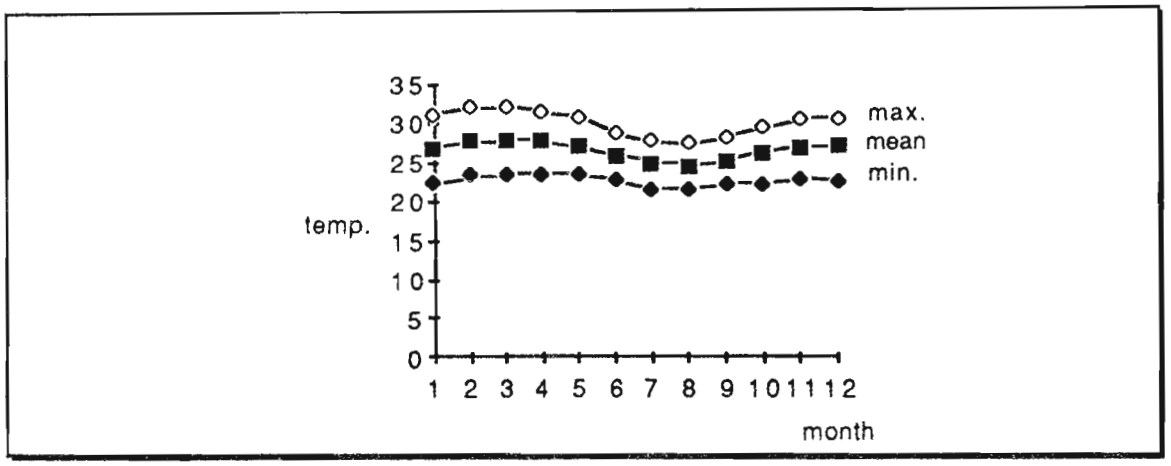


Fig. 1.2. Average monthly temperatures at Adiopodoumé- *Fluctuation annuelle de la température à Adiopodoumé.*

1.2. Aims of the study

The research was intended to provide insight into the role of *B. hancocki* in the transmission and epidemiology of ACMV.

Hitherto no work has ever been done to test the ability of this species to transmit the virus. The capacity of *B. hancocki* to do so is doubted by most authors, but Robertson (1985) indicates that in Kenya in areas with high numbers of *B. hancocki*, incidence of ACMV is also high. Following the establishment of a laboratory culture, transmission tests were designed to determine whether *B. hancocki* is indeed a disease vector while to assess its potential relative importance in disease epidemiology, concurrent field studies of the population dynamics of *B. hancocki* were made.

2. LITERATURE SURVEY ON *BEMISIA* SP.

This part deals with a study on the potential importance of *Bemisia hancocki* as a vector of the African Casava Mosaic Virus (ACMV). In this chapter an outline will be given of the present level of knowledge.

2.1. Geographical distribution and importance of *Bemisia hancocki* (Corbett)

Bemisia hancocki has been recorded from east as well as from west Africa as a pest of cassava *(Manihot esculenta)* (Mound, 1965; Robertson, 1985). The species is known from Southern Europe and Asia to South Africa (Bink-Moenen, 1983). Mound (*loc. cit.*) suggests that *B. hancocki* might originally be African in origin and but which may be gradually being replaced by the exotic *B. tabaci*, introduced probably from Asia. It may be relevant that *B. hancocki* exhibits generation times rather longer than the three to four weeks found locally for *B. tabaci* on cassava (Fishpool, 1988).

Mound (1965) indicates that the rapid spread of ACMV in the first half of this century from east to west Africa could be a reflection of earlier movements by *Bemisia tabaci*. It was found that there was an absence of ACMV from Njala, Sierra Leone in 1960, where then *B. hancocki* alone occurred. This could indicate that *B. hancocki* is not a vector of ACMV. However, Robertson (1987) found that a high incidence of ACMV occurred in Kenyan cassava fields where 40 % of the whitefly population consisted of *B. hancocki*. Hence he suggests that *B. hancocki* might well be a vector of ACMV.

2.2. Identification of *Bemisia hancocki* (Corbett) with reference to *Bemisia tabaci* (Gennadius)

The classification of *B. hancocki* (Corbett) is, as are all members of the subfamily Aleyrodinæ, entirely based upon morphological characters of the so-called pupal case - actually the exuvium of the last instar nymph. This species can easily be confused with *B. tabaci* (Genn.) partially due to the great variability in shape and structure of the pupal case of the latter. Unfortunately there is at present no known way of seperating adults of the two species. The adults of *B. hancocki* may be rather bigger than those of *B. tabaci* (Robertson, 1985) but otherwise they do not possess distinguishable features.

The variation in the pupal case of *B. tabaci* affects mainly the number, length and shape of the setæ, the sculpturing of the dorsal surface and shape of the margin. This variation may be caused by features of the host plant, in particular the degree of pilosity of the leaves, but can also occur within a population on one plant (Mound, 1965; Bink-Moenen, 1983). However, specific differences in the length of the caudal setæ and the shape of the vasiform orifice, although themselves variable in both species, do provide reliable diagnostic characters, and were used successfully in making determinations (see Fig. 2.1).

The vasiform orifice in *B. hancocki* is of an elongate triangular shape with a slight median constriction. The posterior-lateral margins bear some four internally projecting ridges. The opening at the distal

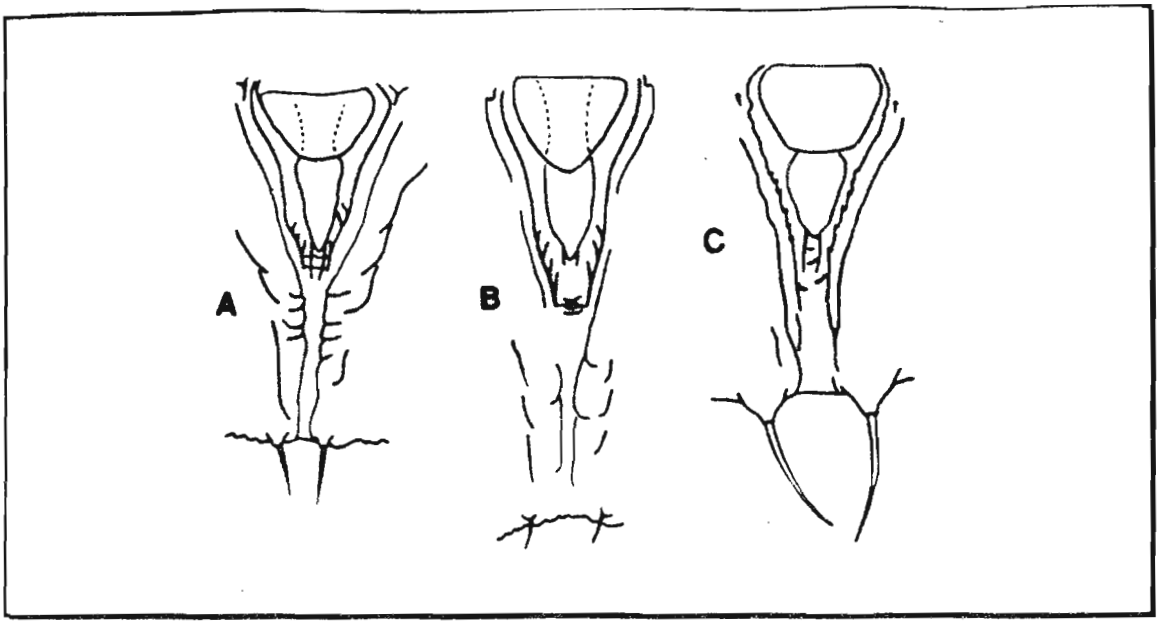


Fig. 2.1. A,B: *Bemisia hancocki* - vasiform orifice C: *Bemisia tabaci* - vasiform orifice (from Mound, 1965)

end is relatively wide. It is 0.07 to 0.11 mm in length, and 0.05 to 0.07 mm in width. The operculum is sub-cordate, filling about half the orifice. The lingula tip is highly variable in shape. It can be conical to elongate triangular or spatulate. It is poorly separated from the lingula neck. The caudal furrow is well defined, 0.05 to 0.09 mm long with 1 or more traverse ridges. The caudal setæ are less than half the length of the caudal furrow, (Mound, 1965), but can be longer on certain host plants (Bink-Moenen, 1983).

The vasiform orifice of *B. tabaci* lacks the transverse markings. It has roughly the same dimensions as the vasiform orifice the *B. hancocki* while the caudal setæ of *B. tabaci* are more than half the length of the caudal furrow (Mound, 1965).

In this research a few additional features of *B. hancocki*, besides the length of the caudal setæ and the shape of the vasiform orifice, were used in making field determinations:

- -shiny appearance, whereas *B.tabaci* nymphs appear matt green-yellow in colour.
- the larger overall size of late instars compared to *B. tabaci* last instars and pupae.
- the rugose dorsal surface, whereas on cassava *B. tabaci* has a smooth dorsal integument.
- The shape of the empty pupal case from which the adult has emerged.

(Fig. 2.2.).

Considerable taxonomic confusion has existed within the genus *Bemisia*. The variation in morphology has lead to a great many species being described, many of which have since been synonymised. Although *Bemisia hancocki* (Corbett) itself has recently been made a synonym of *Bemisia afer* (Priesner & Hosney) by Bink-Moenen (1983), in this report we continue to use the name *B. hancocki* by which it is referred to in the literature, e.g. Mound, (1965); Robertson, (1985) (see Table 2.1).

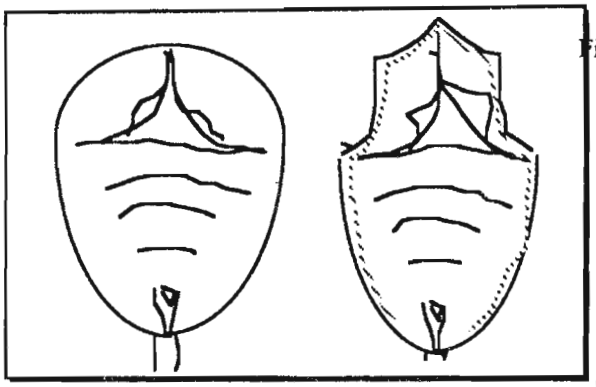


Fig. 2.2. The difference in appearance between the empty pupal cases of *Bemisia tabaci* (left) and *B. hancocki* on the leaf (pers. obs.) - *Difference entre les cages pupales vides de* Bemisia tabaci *(à gauche) et* B. hancocki *sur une feuille de manioc (obs. pers.)*

Species	Synonym
Bemisia hancocki (Corbett)	*Bemisia afer* (Priesner& Hosney) *Dialeurodides afer* (Priesner & Hosney)
Bemisia tabaci (Gennadius)	*Aleurodes tabaci* (Gennadius) *Aleyrodes inconspicua* (Qu.) *Bemisia goldingi* (Corbett) *Bemisia gossypiperda* var. *mosaicivectura* (Ghesquière) *Bemisia longispina* (Priesner & Hosney) *Bemisia nigeriensis* (Corbett) *Bemisia rhodesiaensis* (Corbett)

Table 2.1 Synonyms of *B. tabaci* and *B. hancocki* in Africa- *Synonymes de* B. tabaci *et* B. hancocki en afrique (Mound, 1965; Bink-Moenen 1983).

2.3. Biology of *Bemisia* spp.

Since almost nothing has been published on the life cycle and development of *B. hancocki*, the life cycle of *B. tabaci* is briefly described below.

The genus *Bemisia* belongs to the the Family Aleyrodidae, Order Homoptera. These insects do not possess a complete metamorphosis, but undergo a hemimetabolous development. The immature stages do not resemble the adult insects however, being mainly sessile and morphologically degenerate, but there is no true pupal stage. Although the terms larvae and pupae are frequently used in the literature, they are more correctly termed nymphs (Lopez-Avila 1986, van Helden & van Halder, 1986; Fishpool, 1988).

Bemisia tabaci has four nymphal instars. The newly hatched nymph is about 0,25 mm long and 0,15 mm in width. It is the only nymphal instar which possesses well developed legs. It can move over short distances in order to find a suitable feeding place. It is often referred to as the crawler stage. This is the stage where highest mortality occurs, probably caused by the nymphs falling off the leaves during windy conditions or when handled, (Robertson, 1985). Once the crawler has settled, it ceases all further displacements. It is narrowly eliptical in shape and of a shiny whitish green colour and has a pair of conspicious green organs, the mycetomes, two-thirds of the way down the body. These contain symbionts, important in food digestion. The nymphs were considered to be dead when the mycetomes showed an orange colour (Fishpool, pers. comm.).

The succeeding three instars are sessile. They resemble each other very closely, differing mainly in size. The fourth instar has two distinct eyespots at the anterior end and greatly reduced legs. Up to 8 pairs of setæ are evident, of which

the caudal pair are always present and the most conspicious. The nymphs during these stages are a greenish yellow colour and the mycetomes are large and bright lime green.

The so called pupa is in fact the latter stage of the fourth instar, during which conspicuous morphological changes occur, has a relatively inflated appearance. The colour changes to opaque egg yellow. The segmentation of the abdomen becomes more visible and the red eyespots of the developing adult enlarge and become visible through the nymphal integument. Hence the name 'Red Eyed Nymph'. The nymph is

EGG	7	DAYS
FIRST INSTAR	2.8 ± 0.2	
SECOND	2.8 ± 0.1	
THIRD	2-7	
FOURTH	2-3	
PUPA	2-4	
	19-27	DAYS

Table 2.2 The approximate duration per instar of *B. tabaci* in days - *la durée de vie approxomative du développement des stades larvales de* B. tabaci *en jours.* (Lopez- Avila, 1986)

at this stage 0.5 to 0.75 mm in length and 0.35 to 0.5 mm in width. Some authors state that larger nymphs give rise to females and the smaller ones to males. (Robertson, 1985; Fishpool, 1988).

Last instar nymphs of *B. hancocki* are much less conspicuously coloured. They appear larger and flatter than do last instar *B. tabaci*. The 'pupa' does not swell nor change colour until the adult can be seen through the 'pupal' case. The 'pupa' is 0.67 to 0.98 mm in length and 0.45 to 0.72 mm in width. There is reportedly no sexual dimorphism (Mound, 1965).

Temperature seems to play an important role in the duration of the various stages of *B. tabaci*. The development of the eggs is speeded up from 22.5 days at 16.7°C to 7.6 days at 25°C and 5.0 days at 32.5 °C. They fail to hatch at 36°C. Fecundity decreases a little with increasing temperature, but the percentage of eggs hatching increases over the range 26° to 32°C (Lopez-Avila, 1986).

The duration of the complete life cycle of *B. tabaci* is 19 to 27 days under favourable conditions (Table 2.2.) (Lopez-Avila, 1986). It is not certain how long the duration of the life cycle of *B. hancocki* is in the field. Clear, however, is the fact that *B. hancocki* is differently distributed on the plant, probably due to a longer developmental period (Fishpool, 1988).

2.4. Population dynamics

The adult whiteflies deposit their eggs mainly on the undersurfaces of the upper leaves of the plant. This causes a more or less defined stratification of the different instars within the plant, due to the slower development of the nymphs compared to the development of the new leaves.

Population sizes depend largely on bio-climatological factors such as temperature, precipitation, wind velocity and plant age. It was observed that with the onset of the dry season (December - April) in Ivory coast, whitefly population levels rose and then sharply declined with the return of the rains in May, with which come higher temperatures and relative humidities, (Fig. 1.1. and Fig. 1.2.).

Adult whiteflies are ready, though relatively weak, fliers; they are carried downwind in anything but the lowest windspeeds. The dominant local wind direction is south-west. Newly sprouting cassava fields are very rapidly colonised by immigrants. Van Helden & van Halder (1986) showed that adult whiteflies tend however to move upwind through a cassava crop when the wind velocities are low enough. This, plus the probable accumulation of wind-borne immigrants on the upwind field border in a maturing cassava crop, contributes to the within field clustering of adult *B. tabaci* towards the upwind border, (Fig. 2.3.).

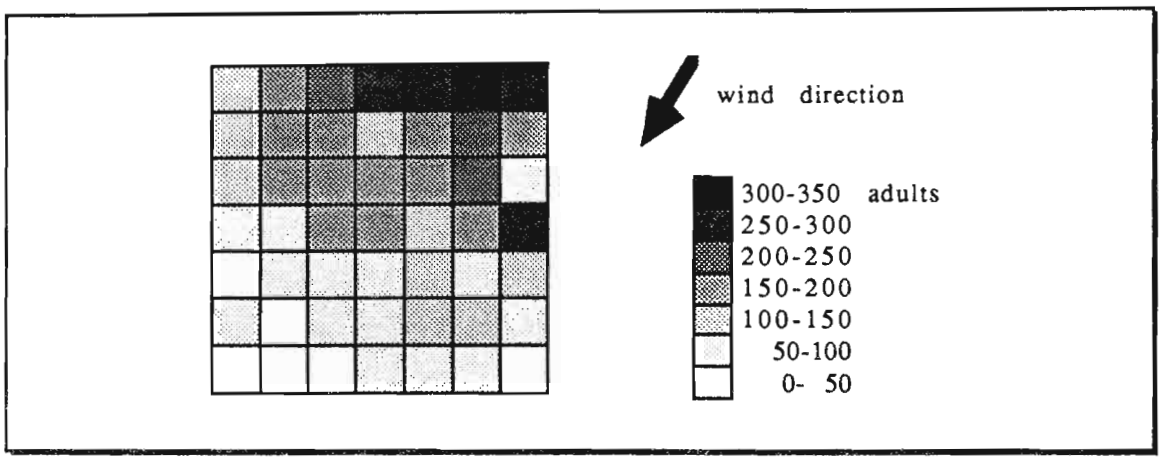


Fig. 2.3. The distribution of adult *B. tabaci* in a cassava field on 26/1/1986- *la distrtibution des adultes de* B. tabaci *dans un champ de manioc le 26/1/1986* (van Helden & van Halder, 1986)

2.5. The transmission of ACMV by *Bemisia hancocki*

Almost nothing is known about the transmission of ACMV by *B. hancocki*. Since this species is closely related to *B. tabaci*, the literature available on the latter will be used as source material.

The success of the virus depends on the behaviour of the whitefly. As only 0.45% of the natural whitefly population is capable of transmitting the virus, it is obvious that a high population in the field is required to give high transmission rates (Fargette, 1987). Whiteflies are attracted by the colour yellow. This helps to explain why cassava cultivars with yellowish petioles bear larger whitefly populations, and hence be more heavily infected than plants with darker petioles (Leuschner, 1977; Colon, 1984).

The virus appears to be transmitted only through young, unexpanded leaves (Chant, 1958; Robertson, 1987). These are exactly the leaves on which adult whitefly preferentially feed and deposit their eggs.

Several studies have been made of the efficiency of the transmission of ACMV. All indicate that very low numbers of viruliferous whiteflies can, within a very short time span transmit the virus. Chant (1958) found that the virus could be transmitted only after an acquisitive feed on diseased tissue of at least four hours. The virus can be transmitted to a healthy plant in a feed of 1/4 hr (see Fig. 2.4.). The number of plants infected increases with increasing feeding time. A plant can be infected by a single fly. However a greater number of whiteflies rapidly increased the transmission rate. An optimum infection rate was acheived using between 30 and 40 adult whiteflies per plant. These figures are

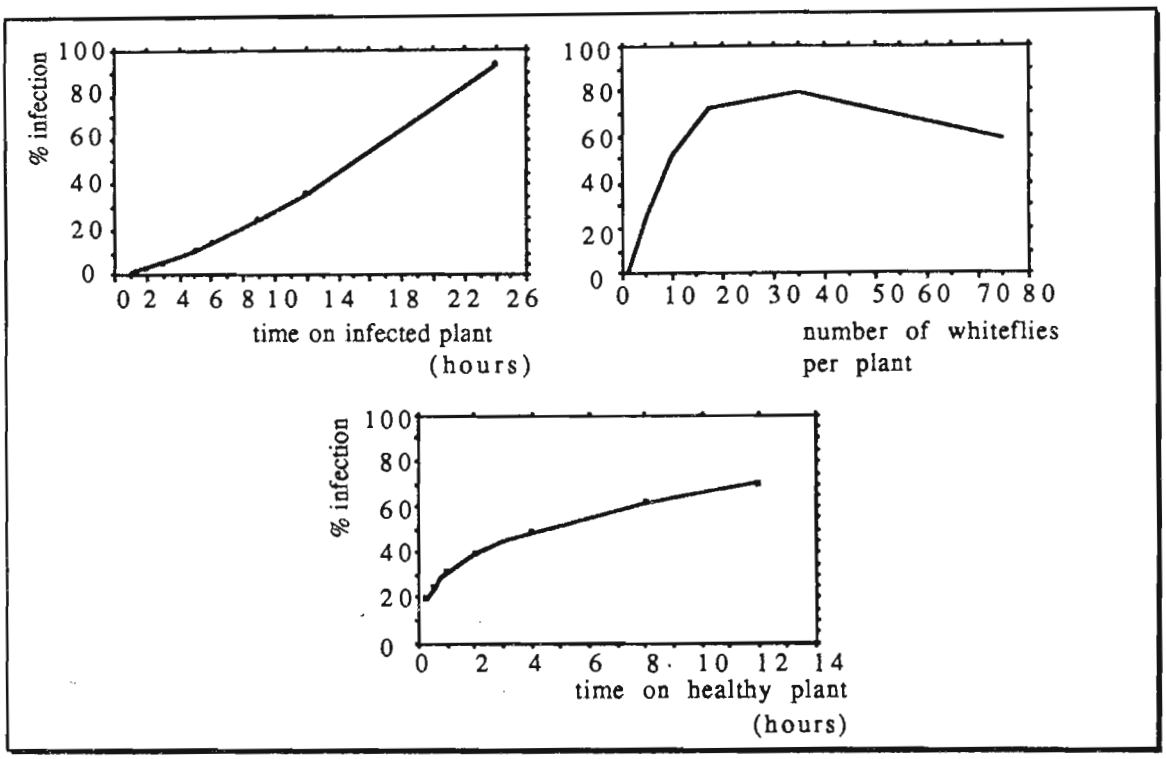


Fig. 2.4. Relationships between feed duration on contaminated cassava by *Bemisia tabaci* and virus acquisition; the number of viruliferous *B. tabaci* and infection of healthy plants and the duration of the inoculatory feed and percentage infecton. - *Relations calculées entre le temps d'acquisition, le nombre de* B. tabaci *viruliféres et le temps d'inoculation et le pourcentage des plantes infectés* (Figures from Chant ,1958).

confirmed by Mahto & Sinha (1978), who found an optimum with populations of approximately 20 individuals per plant. Chant (1958) indicates an incubation time of up to 4 hours, whereas Mahto & Sinha (1978) reported an incubation time of only two hours.

It is possible for the nymphs to acquire the virus and give rise to viruliferous adults; virus is not lost during the metamorphosis. The retention period of the virus is estimated to be 7 to 9 days. Trans-ovarial transmission of the virus is not possible (Dubern, 1979).

Mechanical transmission of the virus has never been demonstrated. Extensive tests have shown that whiteflies are the only vectors and, together with the planting of contaminated cuttings, the only means of transmitting the virus (Dubern, 1979).

3. MATERIALS AND METHODS

The research comprised 3 parts: 1) Field counts of *Bemisia hancocki* in a young disease-free cassava crop to assess field population levels and their variation with time. These were complemented with detailed destructive laboratory counts. 2) An attempt was made to establish a culture of *B. hancocki*, to provide a supply of guaranteed virus free insect material. 3) Transmission tests were done to determine the ability of *B. hancocki* to transmit ACMV.

3.1. The counts

The field counts were carried out over the period from 4 February to 15 June, in two different plots. On 4/2, 11/2, 18/2, 25/2, 3/3, 10/3 , 24/3, 13/4 and 10/5; the nymphal whitefly populations on 13 plants in the first plot were counted, and on 23/3, 30/3, 6/4, 13/4, 20/4, 28/4, 5/5, 12/5, 18/5, 25/5 and 3/6 10 plants from the second plot were similarly treated.

In all counts the leaves were numbered from the uppermost, partially opened (but still red coloured) leaf. This leaf was called leaf n° 1. The next, almost expanded leaf became leaf n° 2, and so on (Fig. 3.1.). Fallen or dry leaves were not counted. Their numbers were omitted.

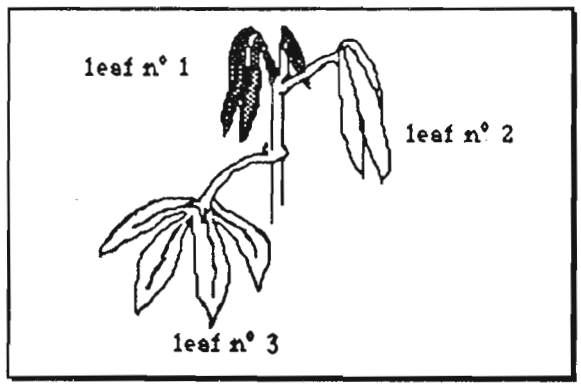


Fig. 3.1. Numbering of the cassava leaves- *Numérotation de feuilles de manioc*

3.1.1. First field

The first field was planted on 26 November 1987. It was also being used in another study, on the population dynamics of *B. tabaci* and its predators. To this end several attractive and non-attractive insect traps, as well as climatological equipment were installed in the field (see Fig. 3.2.).

The field was planted as a square of 49 plots consisting of 100 plants each. The cassava cultivar used was Kasimbidgi Green (Kenyan), which was chosen because of its erect growth and its resistance to ACMV. This cultivar has dark petioles, which may make it initially less attractive to whitefly; however earlier investigations had shown it to support the highest whitefly populations of all the disease-free cultivars locally available, (Fishpool, pers.comm.)

13 plots within the field were selected. In each plot the fifth plant in the fifth row was selected to be monitored. At a suggestion of Dr. L. Fishpool plots were chosen along the south-west field border, as they were considered likely to harbour the largest whitefly populations at the time, and a line of plots at right angles to the first down the middle of the field, from south-west to north-east (Fig. 3.2.).

Each plant had the leaf 10 marked, so that plant growth relative to leaf number could be followed. Counts were made of the number of *B. hancocki* nymphs, the number of fungus-attacked *B. hancocki* and *B. tabaci* nymphs; the number of

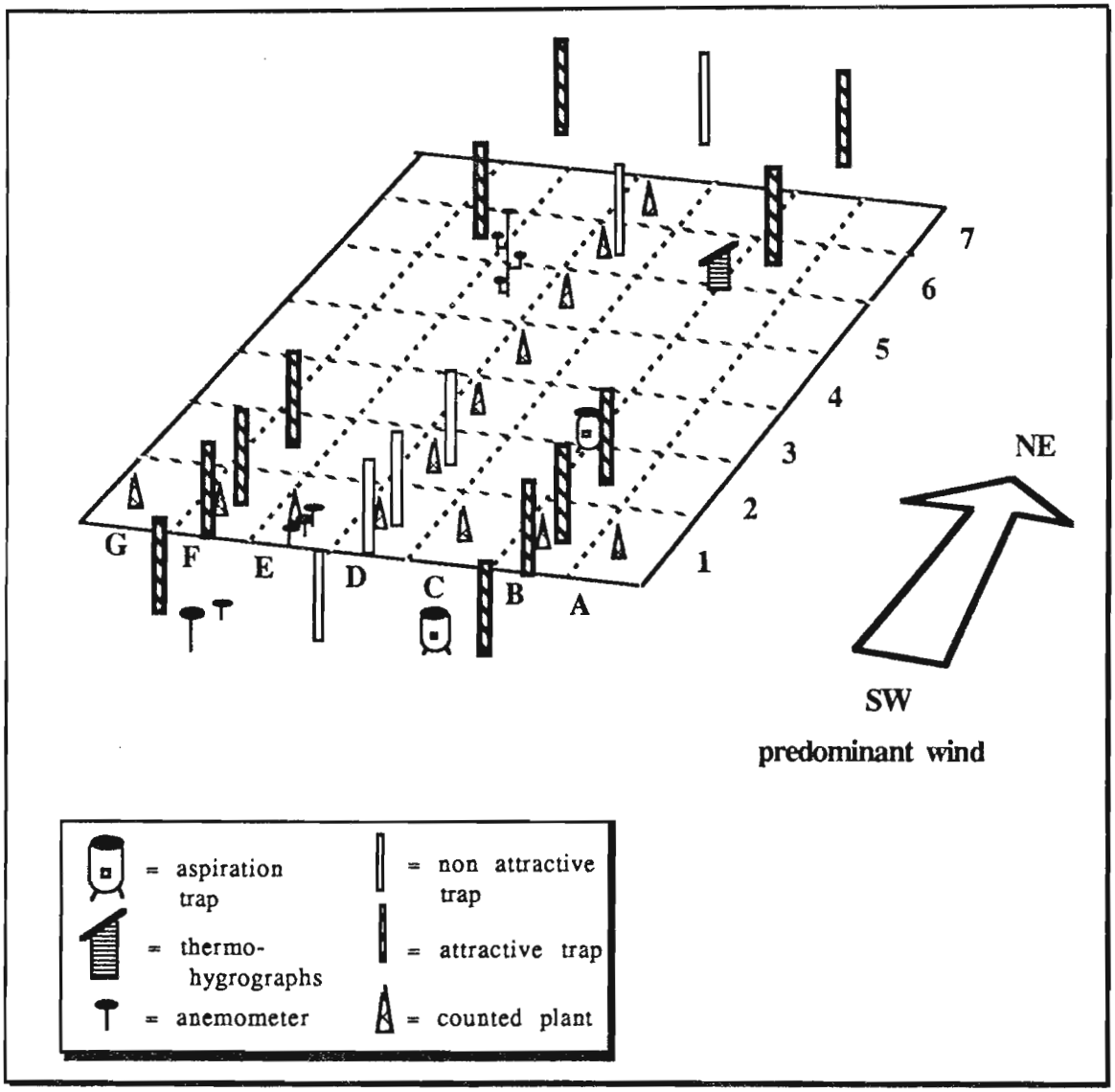


Fig. 3.2. The first field with all traps and equipment shown *in situ*. The sub-plot numbering system and the monitored plants are indicated. - *Le premier champ avec tous les pièges et toute apparature mis en place. La numérotation et les plantes comptées sont indiquées.*

parasitized pupæ of both species, and the number of whitefly predators. All numbers were recorded per leaf. The numbers of *B. tabaci* nymphs were monitored by Dr. L. Fishpool in the same plots, but on other plants. On the last two sampling dates, only the numbers of *B. hancocki* nymphs and coccinellids were counted. The present report considers the *B. hancocki* and *B. tabaci* nymph data only. The parasite and predator results are discussed in a separate report.

3.1.2. Second field

The second field was situated near the north-east border of the first. It was planted on 11 February 1988. The cultivar used was also Kasimbidgi Green, enabling direct comparison of the results with those from the first field.

It was of rectangular shape: 15 rows of 10 plants each. The longest borders of the field were orientated from west to east. The field was divided into ten rectangular plots of 10 plants each, with a border plot of three rows on the eastern side and two border rows tothe west (Fig. 3.3.). In each plot one plant was selected at random using a random number table.

The tenth leaf of the chosen plants were labelled on 23 March. If the tenth leaf was missing, the position of the node was marked. It was thus possible to follow plant growth relative to leaf number as before. The number of *B. hancocki* and *B. tabaci* fourth instar nymphs, the number of parasitized pupæ, the number of predators and the number of fungus-attacked nymphs were all counted over the whole plant. All numbers were recorded per leaf.

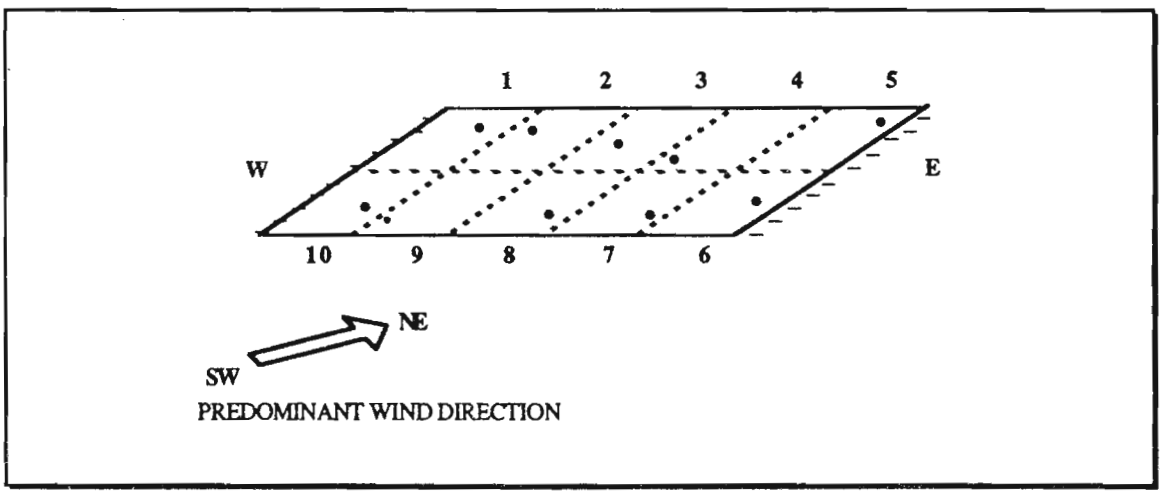


Fig. 3.3. The layout of the second cassava field. The counted plants are indicated with (•) - *Le plan du champ de manioc n° 2. Les plantes suivées sont indiquées par une (•).*

3.1.3. Destructive counts

Destructive counts were undertaken on plants taken from a third field. This field had been planted on 26 November 1987, the same date as the first field. The cultivar used was also Kasimbidgi Green, thus permitting direct comparisons. The field consisted of 20 rows of 10 plants. Plants to be sampled were taken at random from the field. They were transferred to the laboratory, where the leaves were removed

and put in numbered plastic bags. They were stored in a refrigerator until required; in this way both leaves and nymphs could be kept in good condition for at least a week.

Plants to be sampled were gathered on 28/1; 6/2; 9/2; 17/2; 1/3; 30/ 3; 20/4; 5/5 and 18/5.

The leaves were examined under binocular microscopes fitted with eye-piece graticules and the whitefly nymphs counted and measured. In this way it was possible to distinguish between all instars of *B. tabaci* and, for the older instars, to seperate *B. hancocki* from *B. tabaci* and to determine the sex of last instar *B. tabaci* nymphs by their size. It was also possible to infer causes of death of some of the nymphs (parasitised, attacked by fungus or pierced and sucked dry by arthropod predators).

The following data were collected:
- the number of *B. tabaci* :
  - -eggs
  - -first instars
  - -second instars
  - -third instars
  - -fourth instars
    : male and female
  - -pupæ
    : male and female
  - -parasited pupæ
    : male and female
  - -hatched pupæ
    : male and female
  - -hatched parasitized pupæ
    : male and female
  - -fungus attacked nymphs
    :all instars
  - -mite attacked (sucked dry) nymphs
  - -predaceous mites
- *B. hancocki* :
  - -3rd and 4th instars

In this study only the data for *B. tabaci* and *B. hancocki* is used. The data on parasites and predators etc. is discussed in a separate report (Limberg & van Lingen, 1988).

3.2. The culture

Using the method suggested by Robertson (1985), an attempt was made to establish a pure culture of *Bemisia hancocki*. Young Kasimbidgi Green cuttings were used as plant host. This variety was used, despite the low numbers of *hancocki* nymphs found on it in the field, because Kasimbidgi Green was the only entirely healthy variety available.

The potted cuttings were put in insect proof cages, in turn placed in an insectarium. The plants were watered through a sleeve that could be tied off by means of an elastic cord (Fig. 3.5.).

The whiteflies used to establish the colony were trapped emerging from pupæ, by means of micro cages clipped onto the leaves of plants from the first field (Fig. 3.4.). These pupae were identified as *hancocki* in the field using a hand lens but before the adult was released into the culture the identity of its pupal case was confirmed under the microscope. The culture was started at 16 February 1988.

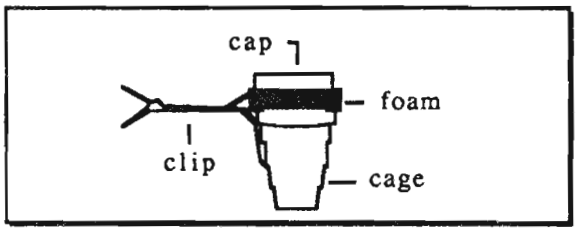


Fig. 3.4. Micro cage- *Cage-micro*

Because of disappointing initial results (see next chapter), it was decided to start four more cultures in smaller cages. These were placed in a CT-room at a temperature of 30 °C and a mean relative humidity of 75 %. The plants were illuminated with fluorescent lamps. In each cage two pairs of adult *B. hancocki* were released. The first pair was released one week before the second. The cultivars used were Kataoli (Togo) and 86 (Ivory Coast). The cuttings originated from the ORSTOM collection at Adiopodoumé and were heavily infected with ACMV. These cultivars were used because of their sensitivity to ACMV and because they were known to capable of supporting good whitefly populations. These cultures were started on 20 April 1988.

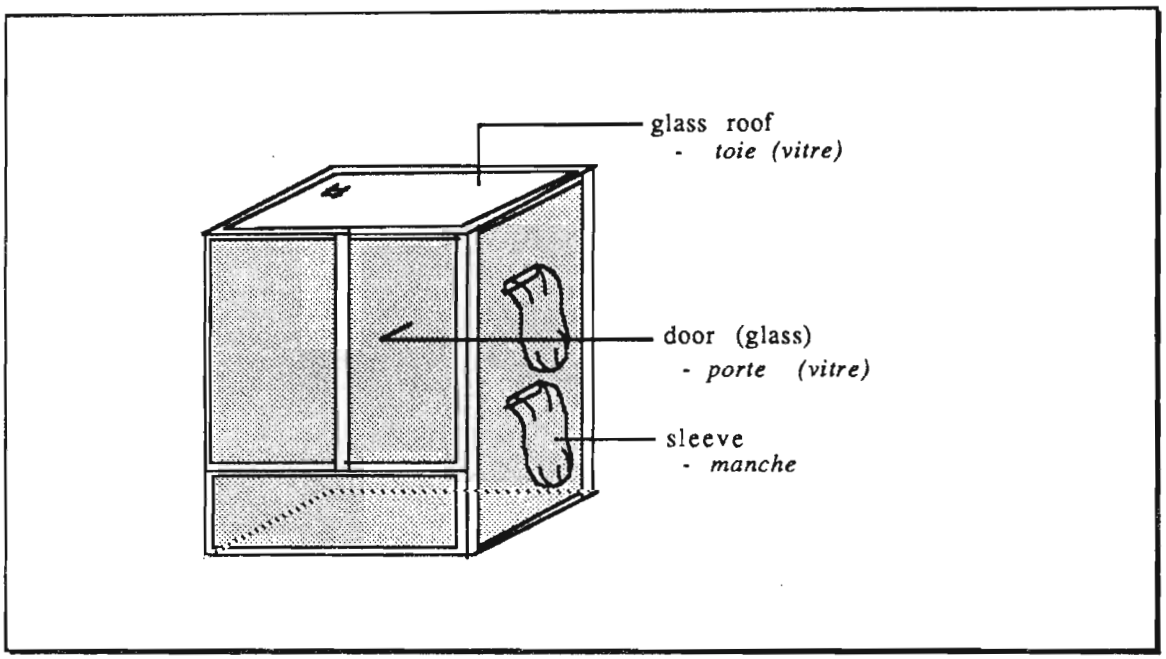


Fig. 3.5. Culture cage ; height = 1,0 m; width = 50 cm; depth = 50 cm - *Cage d'élevage; hauteur = 1,0 m; largeur = 50 cm; profondeur = 50 cm.*

3.3. Transmission of ACMV by *B. hancocki*

To determine whether *B. hancocki* is capable of transmitting ACMV, an experiment was initiated comprising ten replicates and three treatments.

The plant material used in the experiment was cassava cultivar H58 (Madagascar). This cultivar is highly susceptable to ACMV. The cuttings were disease-free and originated from Toumodi, central Ivory Coast, were ORSTOM keeps a limited stock of healthy cassava. The plantation at Toumodi is located in the savanna zone where conditions are less favourable to whiteflies and therefore disease contamination pressure is lower.

Sixty cuttings were placed in a CT-room at a temperature of 30 °C and a mean relative humidity of 75 %. The plants were illuminated with fluorescent lamps. Also three infected plants of the cultivar Kataoli were placed in this CT-room. The CT-room was kept free of whitefly.

Adults of *B. hancocki* were trapped in the field by means of micro cages placed over fourth instar nymphs. Adults of *B. tabaci* were caught using a pooter on plants with high populations of *B. tabaci* nymphs.

The captured adults were placed in micro cages and allowed to feed for 24 hours on the infected Kataoli plants. Thereafter four whiteflies were placed on the young leaves of each healthy H58 plant and left 24 hours to feed. They were then removed from the plants. As controls empty micro cages were placed for 24 hours on the infected plants, after which they were transferred for 24 hours to young leaves on the healthy plants.

The H58 plants were examined after four weeks for symptoms of ACMV.

4. RESULTS

In this chapter the results of the field sampling and the laboratory tests are presented.

4.1. The counts

The first field was sampled regularly from the 70th until the 161st day after planting. The results give an idea of the development and the distribution of the population of *B. hancocki* nymphs within the field.

The second field was regularly sampled from the 41st until the 112th day after planting. In this field the population of *B. tabaci* nymphs was also counted.

The destructive counts were made regularly from the 63rd day until the 176th day after the field was planted. All nymphs were counted.

4.1.1. Development of the plants

During the observation period the growth of the plants was followed by monitoring, as far as possible, the number and position of the leaf nodes.

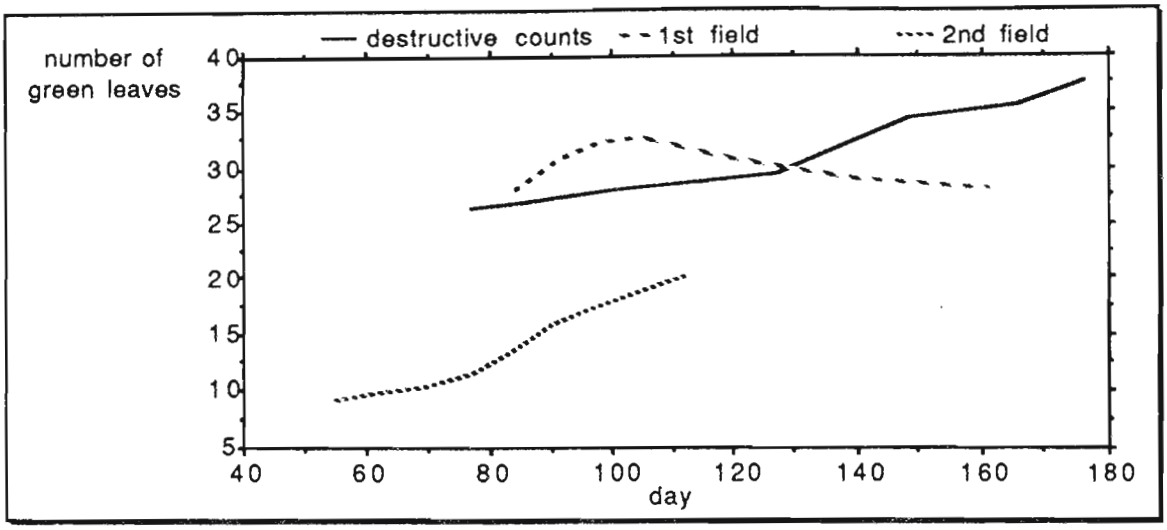
As one can be seen in Table 4.1, the plants in the different fields did not differ significantly in rate of growth. This implies that the speed with which leaves were formed was not substantially affected by weather, or by planting time. Leaf rate formation cannot in this case therefore have had any significant influence on the distributions of the whitefly populations in the plant.

The number of green leaves per plant, however, did differ between the first field and the two smaller fields. The plants in the latter two supported an ever increasing number of healthy leaves, whereas the number of such leaves on plants in the first field reached a peak at about the 105th day (see Fig. 4.1.) and diminished afterwards.

FIRST FIELD			SECOND FIELD		
PLOT	MEAN	T-VALUE	PLOT	MEAN	T-VALUE
A1	5.99	0.852	1	5.02	0.905
B1	3.91	-0.579	2	4.62	0.883
C1	4.30	-0.295	3	4.36	0.276
D1	3.11	-2.89*	4	4.91	0.710
E1	5,50	1,285	5	3,66	-0,522
F1	5,19	0,488	6	3,55	-0,667
G1	6,48	1,190	7	3,12	-5,340
D2	3,50	-3,71*	8	4,49	0,596
D3	2,68	-5,71*	9	3,43	-7,590
D4	3,70	-0,784	10	3,45	-0,770
D5	3,91	-1,012			
D6	5,10	0,729			
D7	5,23	1,078			

	MEAN	TEST MEAN	T-VALUE
FIRST FIELD	4,555	4,08	1,66
SECOND FIELD	4,080	4,55	-7,13

Table 4.1. Mean numbers of cassava leaves produced per plot per week and Student's t-values for differences between the growth rate per plot and the overall field mean, and for differences between fields - *Moyennes et valeurs du T de Student pour les differences entre la croissance par parcelle et la moyenne du champ, et les champs testés entre eux.* * = 5% level ofsignificance.



CORELATION COEFFICIENTS

	DESTRUCTIVE COUNTS	FIRST FIELD**	SECOND FIELD
R	0,979*	0,76*	0,984*
R^2	0,958	0,577	0,968

Fig. 4.1. The mean number of green leaves per plant per field and for the destructive counts. Day = days after planting. - *La numéro moyenne des feuilles vertes par plante par champ et dans la compte destructive. Day = jours après la plantation.*
* = significant at the 5% level
**= 2nd degree polynomial relationship.

4.1.2. Development of the nymphal population

The development of the nymphal populations for the different samples is demonstrated using running means. This is done to overcome the problems posed by large variations between two individual observations. The running means were calculated as the mean of three consecutive observations, e.g. the running mean of day 55 is the mean of the observations of days 41, 48 and 55.

The destructive count data shows a correlation between the age of the crop and the number of nymphs per plant (Fig. 4.2. and Fig. 4.3.). Although the numbers of *B. tabaci* nymphs are much higher than those of *B. hancocki*, the development of the populations largely follows the same pattern. There is a peak, which appears earlier for *B. tabaci* than for *B. hancocki*, followed by a decline, in the number of nymphs. It is noticeable that the decline in the number of living nymphs occurs earlier than the decline in the total number of nymphs for each species. It may be relevant that these declines coincide roughly with the first main rains of the season, (Fig.1.1). The decline may result from physiological changes in the plant, causing adult whitefly to migrate.

Although the number of *B. hancocki* nymphs in the destructive counts declines with time in the same way as the number of *B. tabaci* nymphs does, the proportion of *B. hancocki*

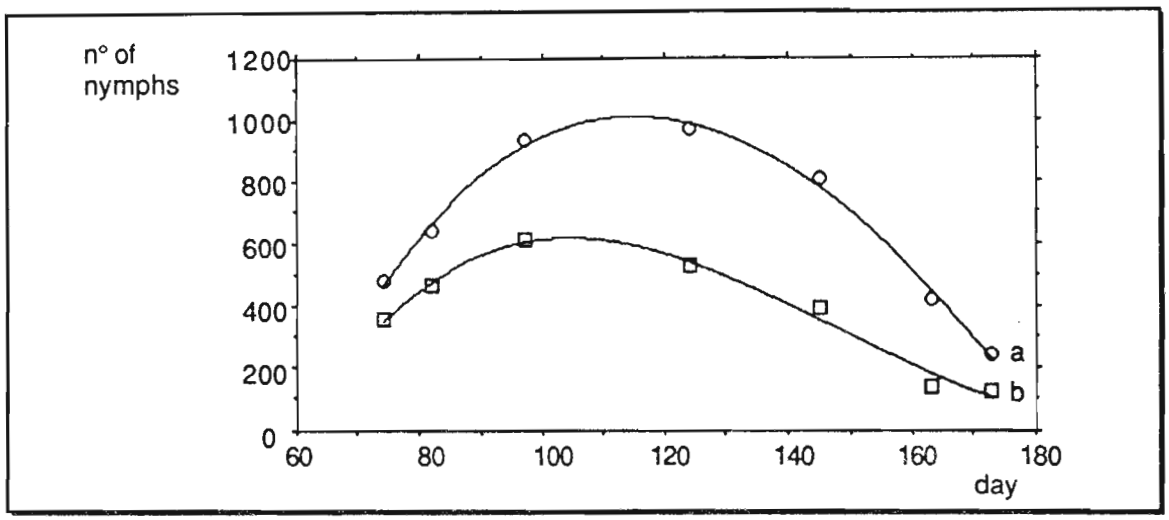


Fig. 4.2. Destructive counts - running means of:
a: total number of *B. tabaci* nymphs per plant
b: number of living *B. tabaci* nymphs per plant ;
day = days after planting
- *Comptes destructifs - moynnes flottantes de:*
a: nombre total des larves de B. tabaci *sur une plante*
b: nombre des larves vivantes de B. tabaci *sur une plante;*
day = jours après la plantation .

formulae: a: $y = -4513,948 + 106,665x - 0,917x^2 + 0,001x^3$ $R^2 = 0,981$
b: $y = -4336,283 + 113,201x - 0,82x^2 + 0,002x^3$ $R^2 = 0,992$

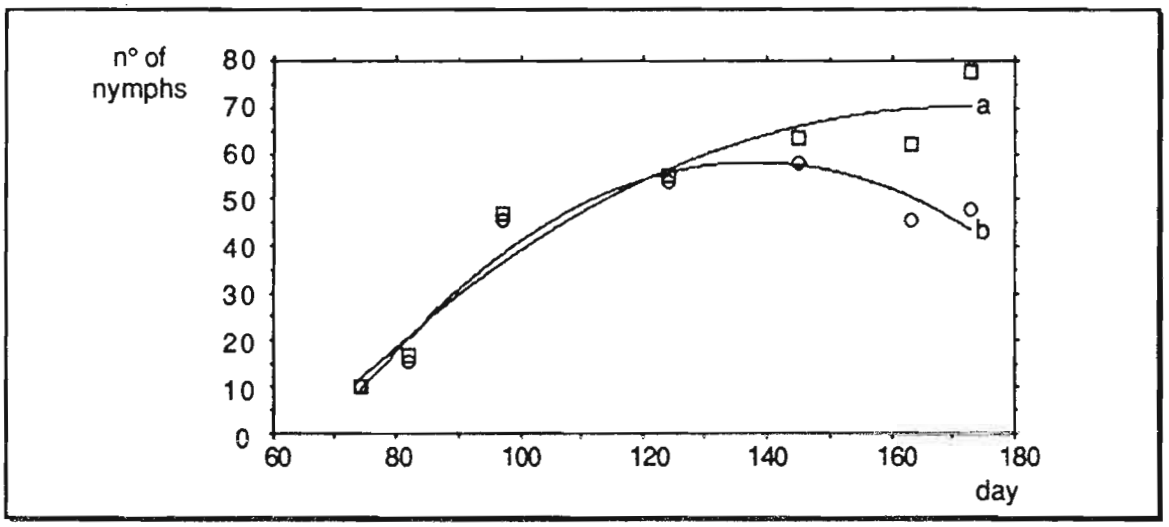


Fig. 4.3. Destructive counts - running means of:
a: total number of *B. hancocki* nymphs per plant
b: number of living *B. hancocki* nymphs per plant
day = days after planting
- *Comptes destructifs - moynnes flottantes de:*
a: nombre total des larves de B. hancocki *sur une plante*
b: nombre des larves vivantes de B. hancocki *sur une plante;*
day = jours après la plantation .

formulae: a: $y = -116,424 + 2,14x - 0,006x^2$ $R^2 = 0,933$
b: $y = -178,513 + 3,357x - 0,012x^2$ $R^2 = 0,939$

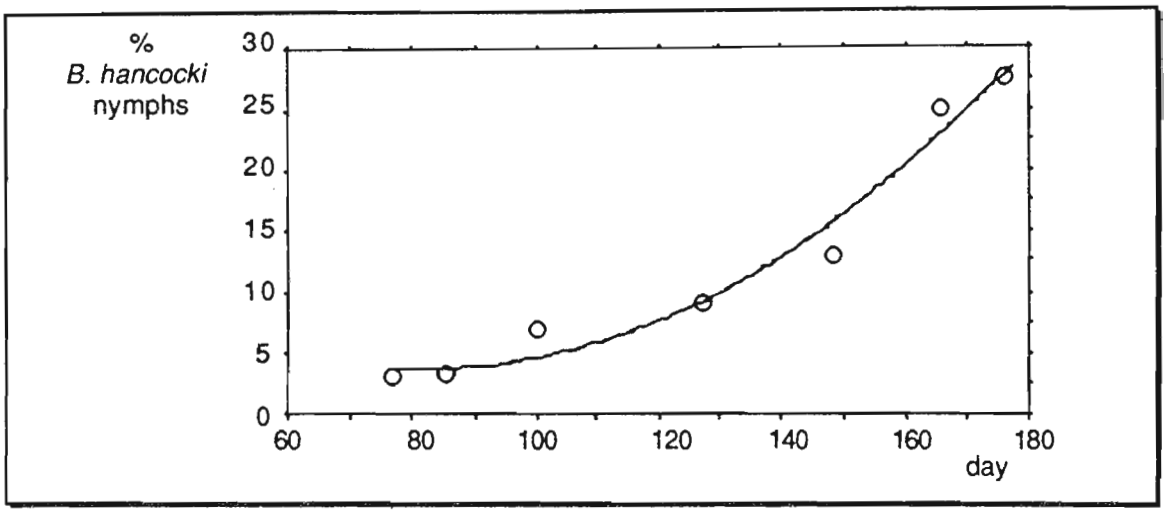


Fig. 4.4. Destructive counts - running means of living *B. hancocki* nymphs as a percentage of the total number of nymphs recorded per plant; day = days after planting - *comptes destructifs - moyennes flottantes du pourcentage de larves de* B. hancocki *vivantes de la population totale sur une plante; day = jours après la plantation .*
formula: $y = 20{,}917 - 0{,}43x - 0{,}003x^2$ $R^2 = 0{,}972$

nymphs of the total living *Bemisia* nymphs increases from 3 to 28 percent during the observation period (See Fig. 4.4.). This results largely from the more rapid decline of the *B. tabaci* population.

The pattern of development of the *B. hancocki* populations in the first field is the opposite of that found in the destructive counts. During the observation period the mean number of *B. hancocki* nymphs steadily declines after an initial slight increase, (Fig 4.5.). The absolute numbers recorded are also lower, but this may be due to the sampling method used (see section 3.1.). Sigificant declines occurred in 5 plots. The other plots also show slight downward trends (Table 4.2.; Appendix A). The differences between these results and those from the destructive samples, taken from the small field, may be related to the size of the fields themselves, as all other (meteorological and agricultural) factors were the same for both. A possible cause of the decline in numbers of nymphs in the first field is the decline with time of the number of healthy leaves per plant, which was observed only there. However, it can be seen from Table 4.3. that only for 6 out of the 13 plots in the first field does a significant relationship exist between mean number of nymphs and mean number of healthy leaves. This relationship is negative in three cases and positive in the others. In addition the overall field mean of nymphal population size does not show a significant relationship with the mean number of healthy leaves. Such a significant relationship, however, does exist for both *B. hancocki* and *B. tabaci* in the two small fields (see Table 4.3.).

During the observation period the number of *B. hancocki* nymphs in the second field remained lower than the numbers observed in the first. This might be due to differences stemming from differing planting dates. The mean population rose during the period of investigation from zero to 3,5 nymphs per plant. The number of *B. tabaci* nymphs also rose in the same period. There was no decline in numbers with time, such as was observed in the first field (Fig. 4.6. and 4.7.). The population per plot showed a significant increase in number, for both *B. hancocki* and for *B. tabaci* (Table 4.2; Appendix B.).

The spatial distribution pattern of the population in the field was not uniform during the observation period. The average nymphal populations per plot are displayed in Fig. 4.8. and Fig. 4.9. It can seen that there is a proportionately large population

CORRELATION COEFFICIENTS, R, OF THE RUNNING MEANS OF NUMBERS OF *B. hancocki* NYMPHS PER PLOT

FIRST FIELD

PLOT	A1	B1	C1	D1	E1	F1	G1	D2	D3	D4	D5	D6	D7
R	-0.08	-0.51	-0.32	-0.59	-0.89*	-0.92*	-0.06	-0.93*	-0.84*	0.27	-0.76*	-0.69	-0.44
R^2	0.01	0.26	0.09	0.34	0.79	0.85	0.04	0.86	0.71	0.08	0.59	0.48	0.19

SECOND FIELD

PLOT	1	2	3	4	5	6	7**	8	9	10
R	0.97*	0.89*	0.45	0.81*	0.66*	0.78*	•	0.81*	0.81*	0.76*
R^2	0.94	0.79	0.17	0.66	0.43	0.61	•	0.65	0.66	0.57

CORRELATION COEFFICIENTS, R, OF THE RUNNING MEANS OF NUMBERS OF *B. tabaci* NYMPHS PER PLOT

SECOND FIELD

PLOT	1	2	3	4	5	6	7	8	9	10
R	0.95*	0.84*	0.81*	0.73*	0.81*	0.97*	0.78*	0.95*	0.81*	0.91*
R^2	0.90	0.71	0.65	0.53	0.66	0.95	0.61	0.89	0.65	0.84

Table 4.2. Corrrelation coefficients of the running means of the number of nymphs per plant per plot with observation dates for the first and second fields.

* : significant at the 5% level

** : no correlation could be calculated.

- *Coefficients de corrélation des moyennes flottantes des numéros des larves sur la plante par parcelle et les date de observation dans le premier et le deuxième champ*

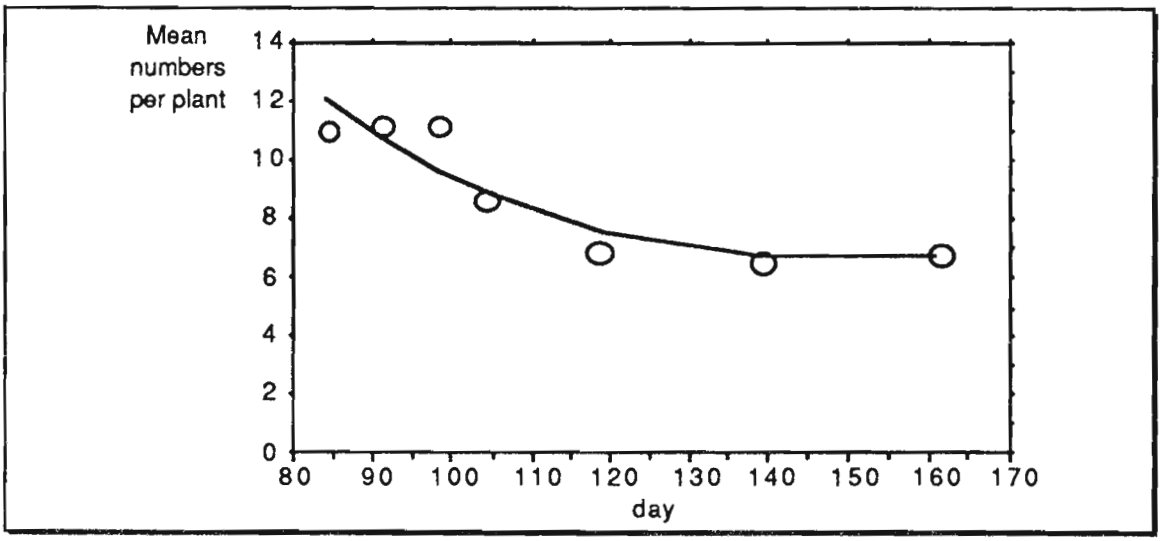


Fig. 4.5. Running means of the average population of *B. hancocki* nymphs per plant in the first field - *moyennes flottantes de la population des larves de* B. hancocki *moyenne par plante dans le premier champ.*

$\ln(y) = 5{,}012 - 0{,}042x + 1{,}388.\ 10^{-4}\ x^2$ $\qquad R^2 = 0{,}879$

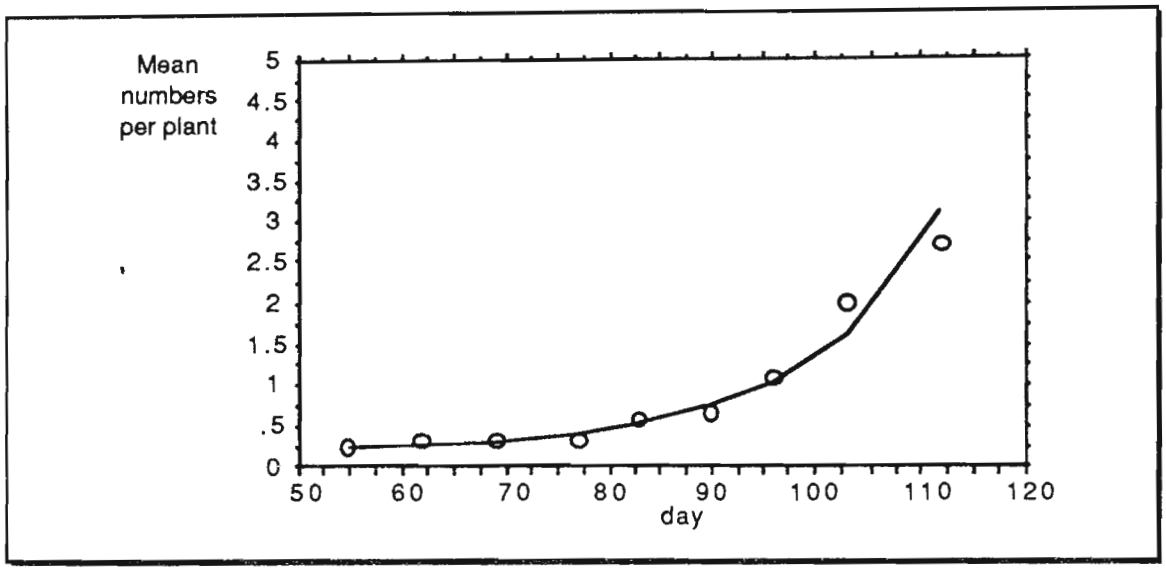


Fig. 4.6. Running means of the average population of *B. hancocki* nymphs per plant in the second field - *moyennes flottantes de la population des larves de* B. hancocki *moyenne par plante dans le deuxième champ.*

$\ln(y) = -0,291 - 0,053x + 0,001x^2$ $R^2 = 0,977$

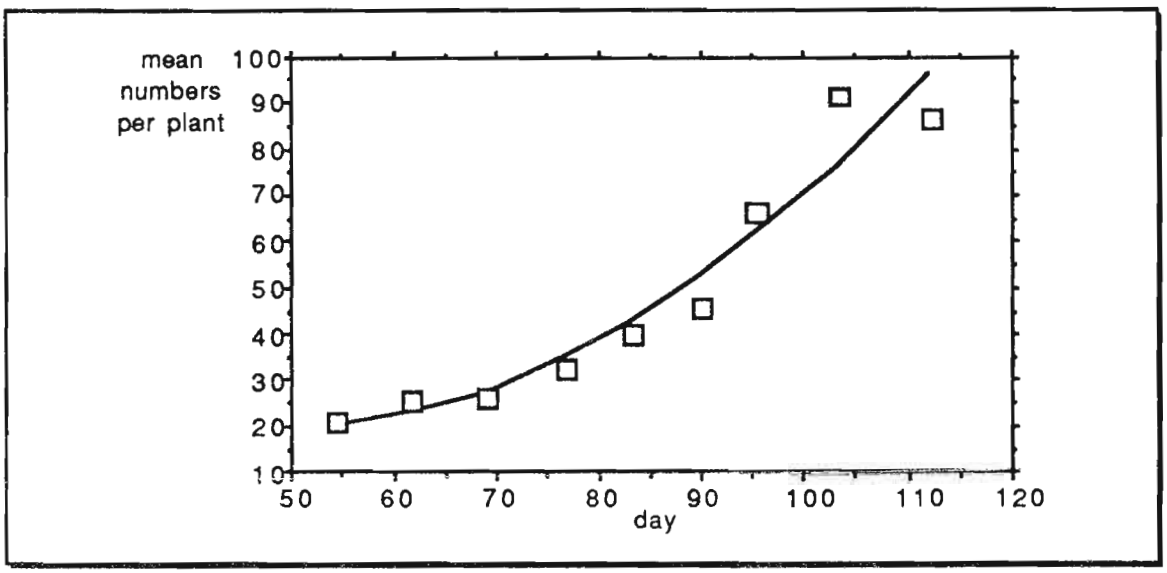


Fig. 4.7. Running means of the average population of *B. tabaci* nymphs per plant in the second field - *moyennes flottantes de la population des larves de* B. tabaci *moyenne par plante dans le deuxième champ.*

$y = 60,09 - 1,739 + 0,018x^2$ $R^2 = 0,933$

DESTRUCTIVE COUNTS

	Bemisia tabaci		*Bemisia hancocki*		TOTAL
	TOTAL	LIVING	TOTAL	LIVING	POPULATION
ΣD^2	78	86	2	18,5	86
ρ	-0,393	-0,536	0,964*	0,67	-0,536

FIELD COUNTS FIRST FIELD *Bemisia hancocki*

PLOT	A1	B1	C1	D1	E1	F1	G1	D2	D3	D4	D5	D6	D7	TOTAL POP.
ΣD^2	29,5	101,5	17	86	110	100	58	2	1	46	12,5	69,5	56,5	32
ρ	0,473	-0,81*	0,696	-0,536	-0,96*	-0,79*	-0,036	0,96*	0,92*	0,179	0,78*	-0,241	-0,009	0,429

FIELD COUNTS SECOND FIELD *Bemisia hancocki*

PLOT	1	2	3	4	5	6	7	8	9	10	TOTAL POP.
ΣD^2	13,5	10,5	98	10,5	77,5	31	59,5	18,5	19,0	24,5	0,5
ρ	0,887*	0,913*	0,183	0,913*	0,354	0,742*	0,504	0,846*	0,842*	0,796*	0,996*

FIELD COUNTS SECOND FIELD *Bemisia tabaci*

PLOT	1	2	3	4	5	6	7	8	9	10	TOTAL POP.
ΣD^2	3,5	28	42,5	56	30,5	4,5	8,5	12,5	38	10	2
ρ	0,971*	0,767*	0,646	0,533	0,746*	0,962*	0,929*	0,896*	0,683*	0,917*	0,983*

Table 4.3. Spearman's rank correlation coefficients between the number of nymphs and number of healthy leaves (all observations in the destructive counts taken as replicates and for the two fields per plot) - *Coefficients de correllation de rang de Spearman pour la relation entre la numéro des larves et la numéro des feuilles vertes (toutes observations des comptes destructives pris comme répétitions et pour les deux champs par parcelle).*

ρ = Spearman's rank correlation coefficient

* = significant at the 5% level.

in plot G1 of the first field. This is in agreement with the results obtained by van Helden and van Halder (1986) (see section 2.4.).The differences between the plot means are only significant in 19 out of 85 cases. The mean of plot G1 differs significantly from all others (Fig 4.10.; Table 4.4.). In addition, plot E1 differs signicantly from plots D1, D2, D3 and D6. Plot F1 shows a significant difference from plots D1, D2 and D3. Plot A1 differs from plots D1 D2 and D3 and plot C1 differs significantly from plot D2.

From this one may observe that in the first field the concentration of *B. hancocki* nymphs was highest in the west-southwestern corner of the field and lowest in the middle of the southwestern border (Fig. 4.8.).

This same phenomenon can also be seen in Appendix C where the distribution patterns of *B. hancocki* in the first field on individual days are shown. On most dates plot G1 supports the largest population. The plots in row D, especially those near the southwestern border, usually have low to very low numbers of nymphs.

In the second field it was not possible to infer any relationship between dominant wind direction and the distribution of *B. hancocki*, due to the very low numbers recorded. Most plants supported an overall average population of less than one nymph per plant, with a maximimum of 3.18 per plant.

The largest population occurred on plant 1, which was on the western border, and thus, with plant 10, the most exposed to the dominant southwesterlies (see Fig. 4.9.). The difference between it and all other plots is significant. Furthermore plant 7, on which no nymphs were recorded

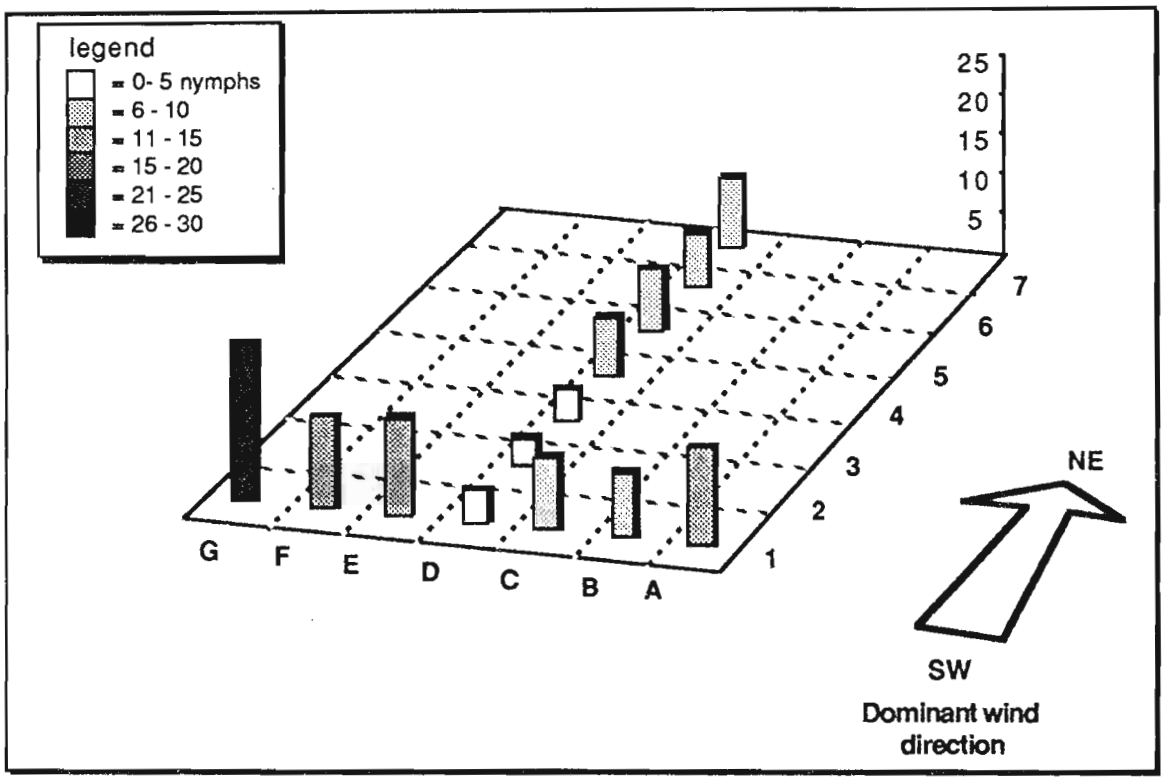


Fig. 4.8. The distribution pattern of mean numbers of *B. hancocki* nymphs during the observation period in the first field - *La distribution moyenne des larves de* B. hancocki *dans le premier champ pendant la période d'observation.*

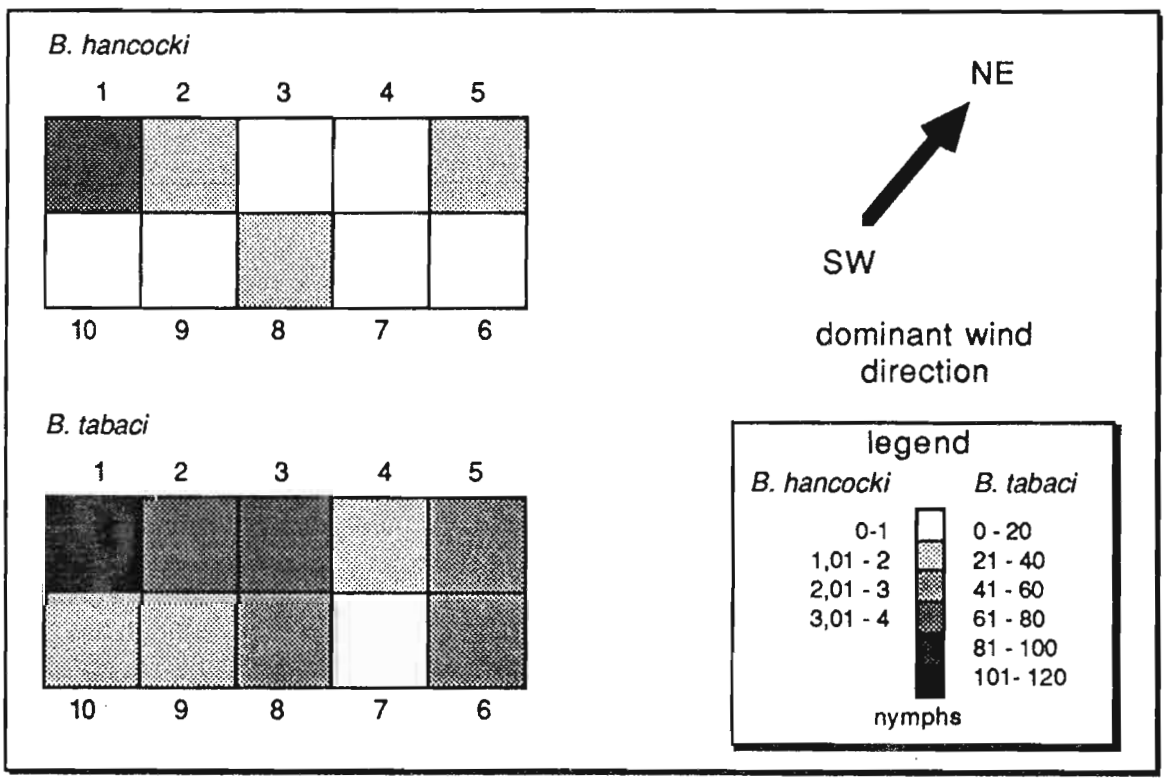


Fig. 4.9. The distribution pattern of mean numbers of *B. hancocki* and *B. tabaci* nymphs in the second field during the observation period. - *la distribution moyenne pendant la période d'observation des larves de* B. hancocki *et* B. tabaci *dans le deuxième champ.*

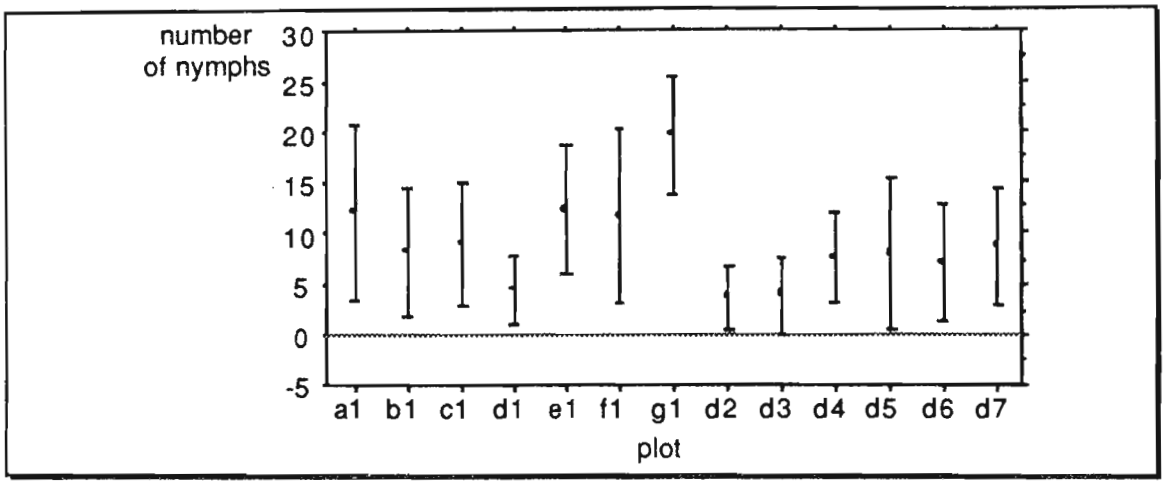


Fig. 4.10. First field - mean numbers of *B. hancocki* per plant with 95% confidance limits - *Premier champ - numéros moyennes de* B.hancocki *par plante avec l'intervalle de confiance à une seuil de 5%.*

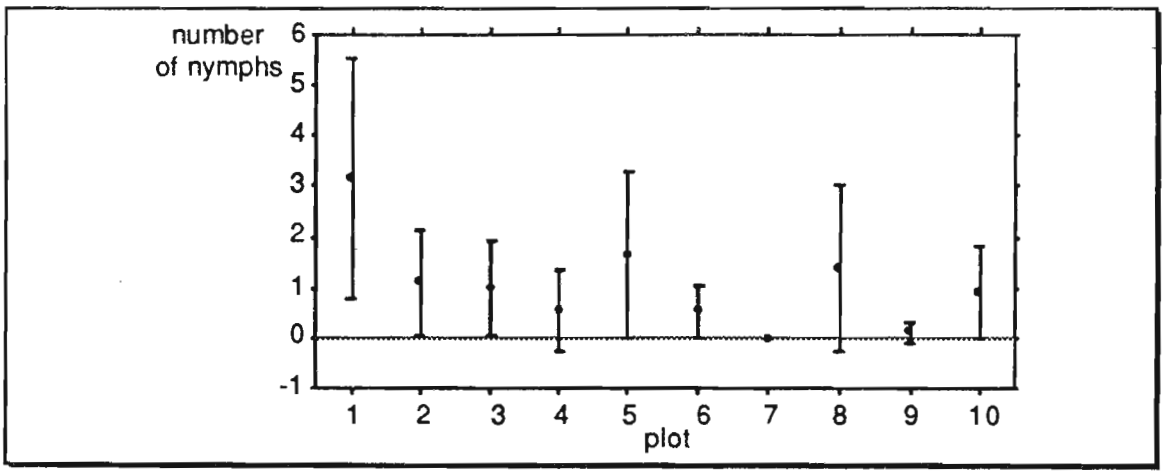


Fig. 4.11. Second field - mean numbers of *B. hancocki* per plant with 95% confidance limits- *Deuxième champ - numéros moyennes de* B.hancocki *par plante avec l'intervalle de confiance à une seuil de 5%.*

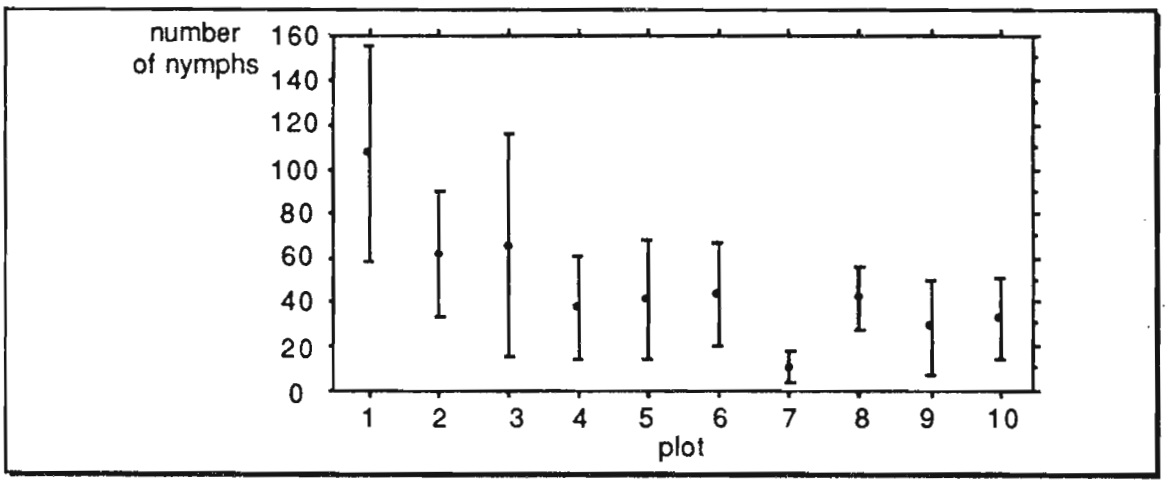


Fig. 4.12. Second field - mean numbers of *B. tabaci* per plant with 95% confidance limits- *Deuxième champ - numéros moyennes de* B.tabaci *par plante avec l'intervalle de confiance à une seuil de 5%.*

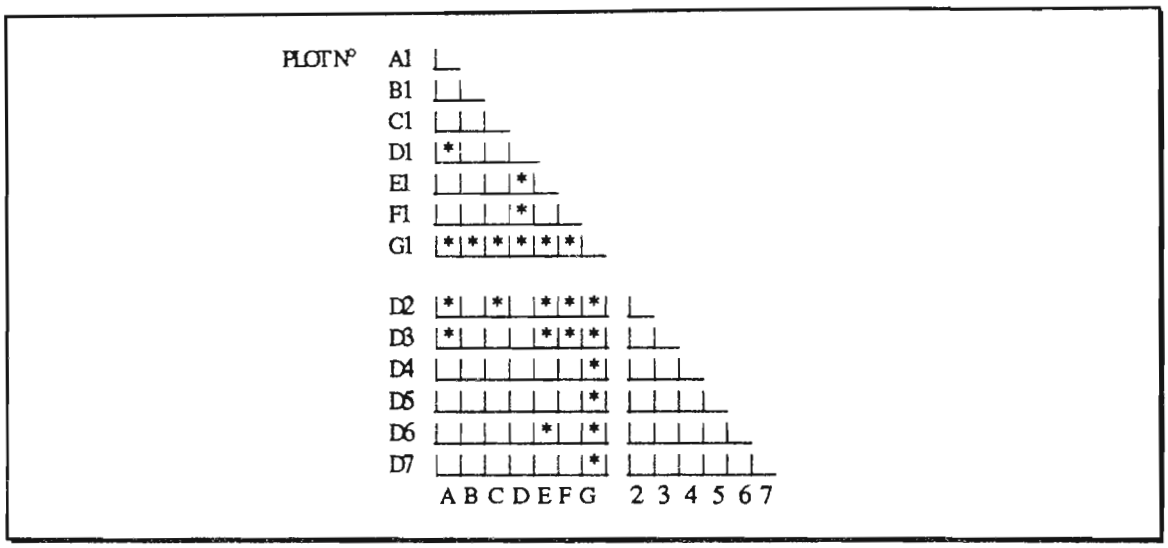


Table 4.4. First field - Significance diagram of Fischer's Protected Least Significant Difference (PLSD) of the mean population per plant during the observation period. - *Premier champ - Diagramme de significance de la Plus Petite Différence Significative (PLSD) de Fischer de la population moyenne par plante pendant la période d'observation.*
B. hancocki: PLSD = 5,246

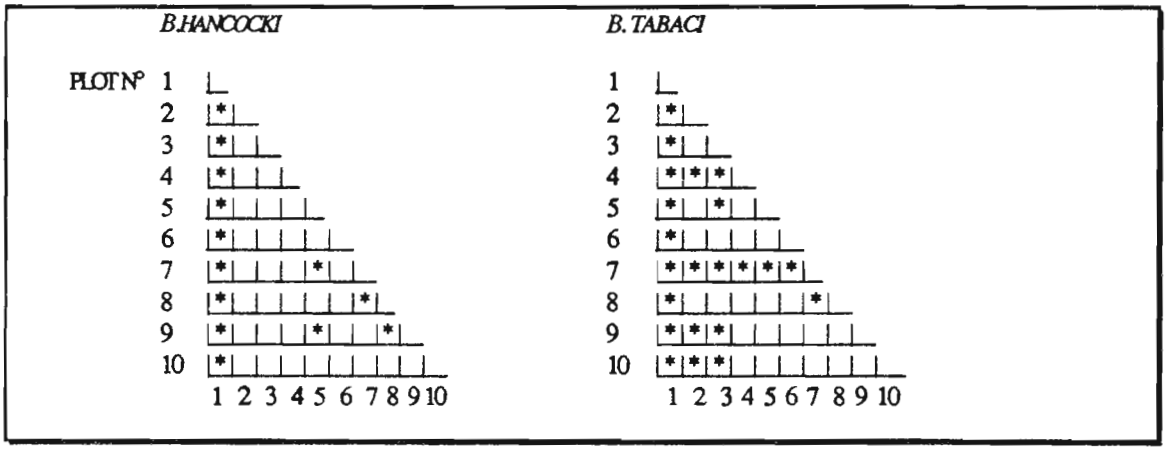


Table 4.5. Second field - Significance diagram of Fischer's Protected Least Significant Difference (PLSD) of the mean population per plant during the observation period. - *Deuxième champ - Diagramme de significance de la Plus Petite Différence Significative (PLSD) de Fischer de la population moyenne par plante pendant la période d'observation.*
B. hancocki: PLSD = 1,216
B. tabaci: PLSD = 23,954

at all, differed significantly from plants 5 and 8. Plant 9, which had supported very few nymphs, also differed significantly from plants 5 and 8 (Fig. 4.11; Table 4.5.).

The population densities of *B. tabaci* nymphs in the second field were also very low, compared to the numbers observed in the destructive counts and in the first field. The pattern of distribution of the population mirrors that observed for *B. hancocki:* Plants 1, 2 and 3 held the highest populations and plant 7 the lowest. These plants differ significantly from those of most of the remainder, (Fig 4.12; Table 4.5.).

The same trends are visible in the observations on particular dates (see Appendix D). Although no more than 9 *B. hancocki* nymphs on a plant was observed, it can be seen that numbers on plant 1 tend to be highest, with plant 7 and plant 9

having the lowest. Other than this no pattern emerges.

The distribution pattern of *B. tabaci* on specific dates shows, not surprisingly, the same pattern as that observed for the mean population. The highest numbers were found on plants 1, 2 and 3. There appears to be a trend to greater numbers towards the western border.

From its relative position it might have been expected that plant 10 would have held the largest number of nymphs; this was what was seen in the first field. That it was found not to do so may be due to the difference in size between the two fields, perhaps thus differently affecting wind flow through and over the fields and therefore the pattern of arrival of downwind migrant adult whiteflies.

4.2. The culture

The size of the *B. hancocki* culture was only assessed on three occasions since it was felt that unnecessary disturbance to the colony should be kept to an absolute minimum.

Between 16 February and 18 May two adult *B. hancocki* were released almost daily into the cage. The release rate was dependant upon the speed of development of the *B. hancocki* nymphs that had been clipcaged in the field. From the average time it took the pupæ to emerge in the field after caging, (>1 week), it was estimated that total nymphal development time would exceed six weeks.

The culture was first counted on 1 April 1988, 6 weeks after it had been established. It was found then that the culture had become heavily contaminated with *B. tabaci* nymphs. Detailed checking revealed that the invaders had introduced themselves, probably during the germination of the cassava in a non insect-proof greenhouse. From this it is at least clear that *B. hancocki* does not have as fast a reproductive rate as *B. tabaci*.

It is clear from Table 4.6 that the population of nymphs diminishes with time instead of showing the expected increase. This might have been caused by a high mortality rate amongst the released adults, perhaps attributable to the high daily temperatures recorded in the insectarium, which occasionally rose above 37 °C (Fig. 4.13). The literature indicates that very high temperatures can have deleterious effects upon whiteflies, (Lopez-Avila 1986 - see section 2.3.).

As a result on 20 April, four new cultures were started on four plants in seperate cages in a CT-room. In each cage two adult *B. hancocki* were released on

	B. hancocki							*B. tabaci*			
	1 APRIL		5 APRIL	11 APRIL		26 MAY		1 APRIL		5APRIL	11APRIL
PLANT	N	H	N	N	H	N	H	N	H	N	N
I	12	9	0	9	3	12	29	5	9	0	1
II	9	0	15	4	0	1	5	23	60	3	0
III	16	9	0	11	12	2	0	1	46	0	1
IV	21	0	3	1	1	0	0	5	83	0	0
TOTAL	58	18	18	25	16	15	24	34	198	3	2

Table 4.6. The development of the culture in the insectarium. - *developpement de la culture dans l'insectarium.*
N = nymphs H = hatched - *émergé*

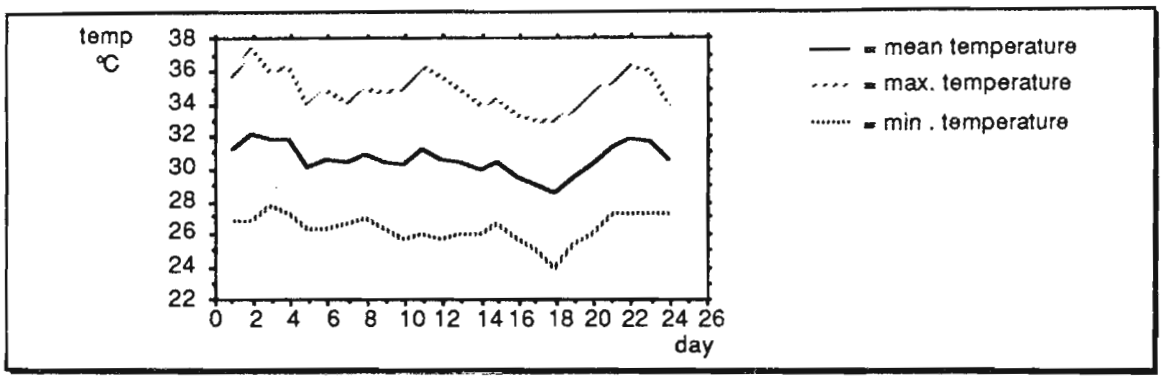


Fig. 4.13. Temperatures in the insectarium from 14 April to 12 May (running means)- *températures dans l'insectarium de 14 Avril à 12 Mai (moyennes flottantes)*

20 April and two more one week later. On 26 May the plants were examined to verify:

a) whether *B. hancocki* can produce nymphs that closely resemble those of *B. tabaci*
b) whether the high temperatures in the insectarium could have been the cause of low reproductive rates.

The temperature in the CT-room varied between 27 °C and 30 °C, which approximates to the average daily temperature in the field.

In the CT-room cultures no *B. tabaci* nymphs were found. Only on one plant was a *B.hancocki* nymph found (Table 4.7.). This could indicate that *B. hancocki* adults suffer high mortalities within the micocages and/or during transferral but this was not borne out by experiences during the transmission tests (see next section). A more likely explanation is that the released *B. hancocki* population contained only a low proportion of females as the adults were not sexed prior to introduction; alternatively those females that were released may have laid few eggs. Which of these explantions, if either, is correct is unknown; it is clear however that the effects of temperature on the fecundity and development of *B. hancocki* requires further study.

PLANT	N°OF NYMPHS
I	0
II	1
III	0
IV	0

Table 4.7. The number of *B. hancocki* nymphs recorded in the CT-room cultures - *le numéro des larves de* B. hancocki *dans les cultures dans la logette climatisée*

4.3. The transmission of ACMV by *B.hancocki*

The transmission tests could be performed twice only, due to the delay caused by the prolonged development of the immature *B. hancocki*. The first test was carried out on 24 June, using three blocks of two test plants with three treatments; one as control, and one each with the two *Bemisia* species. The second test was to have taken place on 4 July. It could not take place however because during a brief unavoidable absence of the investigators the experimental plants dried out.

The results of the first test are shown in Table 4.8. The prescence or absence of ACMV was assessed by prescence or absence of visible symptoms. The only symptoms that could be seen were little chlorotic yellow spots on the newly formed leaves. The characteristic mottling and deformation of the leaves normally caused by the virus were not seen.

Table 4.8. one can be interpreted to

mean that both *B. hancocki* and *B. tabaci* can transmit ACMV. However, it would be unwise to draw too firm conclusions from these observations. More replicates using more plants need to be made before it can be said that the virus can indeed be transmitted by *B. hancocki*. During the test the suspection arose that the reputedly originally healthy clones used might have already been lightly contaminated with ACMV, because of the appearance of some marks on the leaves of the control plants. These marks may also have been caused by phytophagous mites (probably Cassava Green Mite) that had infested the test plants several days before the observation date.

BLOCK		I	II	III
TREATMENT	PLANT			
CONTROL	1	-	-	-
	2	-	-	-
B. tabaci	1	+	+	+
	2	+	+	+
B. hancocki	1	+	+	+
	2	+	+	+

Table 4.8. The presence or abscence of symptoms of ACMV on the experimental plants - *la présence des symptomes du ACMV dans les plantes expérimentales*
- = no symptoms observed- *pas des symptomes*
+ = yellow spots on new leaves - *taches jaunes sur les feuilles neuves*

5. DISCUSSION AND CONCLUSION

In this chapter our results, plus observations published elsewhere will be, as far as possible, drawn together into a conclusion. Some suggestions will also be made concerning future research.

5.1 Discussion

From this study it seems that *B. hancocki* does not occur in large numbers on cassava in southern Ivory Coast. The reasons for this are not known with certainty. It may be that cassava is generally a poor host for *B. hancocki*. Its relative rarity compared with *B. tabaci* is probably at least partly attributable to longer generation times and apparently lower fecundities. That even in a supposedly pure culture of *B. hancocki*, *B.tabaci* was able to invade and swamp *hancocki* is a measure of this. This supports the hypothesis of Mound (1965), that the endemic *B. hancocki* is being out-competed and replaced by the introduced *B. tabaci*.

Due to the low number of *B. hancocki* nymphs found in the field and in the cultures it was not possible to determine accurately the developmental time of *B. hancocki* from egg to adult. However, from our observations the developmental time can be estimated. Let it be assumed, (with what validity is not known,) that the duration of each instar of *B. hancocki* is in the same proportion to the corresponding stage of *B. tabaci*, as that of their pupal stages and that *B. hancocki* has the same number of nymphal instars as *B. tabaci* The time taken for *B. hancocki* pupæ to hatch in the field is 5 to 9 days. As described in section 2.3, *B. tabaci* pupæ hatch 2.5 times more quickly. This would mean that *B. hancocki* takes from 47 to 67 days to develop from egg to adult. Since in the field, plants formed about 4 leaves every 7 days, a *B. hancocki* nymph resulting from an egg deposited on the first leaf, is likely to emerge as adult from between leaves 27 to 43. If true this may also be a cause of high mortality due to the of the dying of the leaf before emergence of the adult. During the observation period average numbers of healthy leaves did not exceed 40 per plant.

From the transmission test one can conclude that there exists a strong indication that *B. hancocki* can transmit ACMV. This supports the observations of Robertson (1987) in Kenya, who observed a strong ACMV contamination of cassava fields that were heavily infested with *B. hancocki* (see section 2.1.).

In both plots studied, the numbers of *B. tabaci* found were far higher than the numbers of *B. hancocki*. The latter, however, seems to react less strongly to the onset of the rains and the aging of the cassava crop. The number of *B. hancocki* as a percentage of the total nymphal population increases steadily as the crop matures. This coincided in two fields with the onset of the rains, so that it is not entirely clear whether the decrease in *B. tabaci* numbers and the corresponding increase of the percentage of *B. hancocki* nymphs was due to the effects of the rains, the lower maximum temperatures or the age of the crop.

The within plant distribution of the *B. hancocki* population did not vary with the growth of the crop, which was in fact fairly steady. This is to be expected, since the adults deposit their eggs mainly on the upper leaves. Therefore, irrespective of how quickly a plant produces new leaves the adults use only a limited number of the available leaves.

The within field distribution of *B. hancocki* resembled that of *B. tabaci*. The highest concentration of nymphs was observed in the southwest corner of the first field. Although the highest concentration of both species in the second field occurred in the northern plots, which were not exposed directly to the prevailing

wind, this may in fact be compatible with the trend seen in the first field, as mentioned in section 4.1.2. This distribution pattern is similar to others observed on this and similar plots and described elsewhere. The peak of numbers in the southeastern corner is noteworthy and had not been seen in the earlier studies on *B. tabaci*. It is without obvious explanation, although the rather poorer growth of the cassava in the central plots, (C1-E1 and D2-D7), due to heavy infestation with Cassava Green Mite, may be relevant. For further details of this, see the report on natural enemies of *B. tabaci* (Limberg & van Lingen, 1988).

Peak numbers of *B. hancocki* occurred in this study about the 140th day after planting. By this time, cassava cultivars sensitive to ACMV would have already become contaminated. Until the 120th day *B. hancocki* did not constitute more than 5 % of the total population of whitefly, of which only some 0.45 % is viruliferous (section 2.5). The highest observed mean number of living *B. hancocki* nymphs was 55 third and fourth instars per plant, which, assuming relatively low subsequent mortality - the literature suggests that most mortality occurs during the first instar, at least for *B. tabaci* - would give rise to approximately the same number of adults. The highest observed mean number of viable *B. tabaci* nymphs was 600 third and fourth instars. This would imply 0.23 viruliferous *B. hancocki* adults developing per plant, against approximately 2.7 viruliferous *B. tabaci* adults per plant over the same period. For these reasons, although *B. hancocki* may well be a vector of ACMV, it is highly unlikely that it is of major epidemiological importance in the coastal region of Ivory Coast.

5.2 Conclusions

From the report one can draw the following conclusions:

- *Bemisia hancocki* does not occur in high numbers in the cassava fields of the coastal region of Ivory Coast.
- The distribution of *Bemisia hancocki* within a cassava field shows the same pattern as has already been shown for *B. tabaci*. The highest concentration of nymphs was observed in the corner of the field that is most directly exposed to the prevailing wind.
- Initially the numbers of *B. hancocki* nymphs in the cassava crop represented only a small fraction of the total nymphal *Bemisia* population but increased rapidly after the 120th day. This is not caused by a significant increase in the size of *B. hancocki* population, but by the rapid decrease of the numbers of *B. tabaci* nymphs.
- The developmental rate of *B. hancocki* is rather slow and its fecundity appears relatively low. The species can easily be replaced by *B. tabaci*, which has a correspondingly higher rate of reproduction.
- It is absolutely essential to protect a culture of *B. hancocki* against all other entomological and phytopathological influences, since the nymphs have to remain on the leaves for at least 6 weeks.
- There is a strong indication that *B. hancocki* can vector ACMV.
- *B. hancocki* is not likely to be an important vector of ACMV in Ivory Coast, because of its low numbers and slow rate of multiplication.

5.3 Recommendations for further research

From this research some practical points emerged. It is our opinion that to monitor the nymphal population of a relatively rare whitefly species such as *B. hancocki,* large fields should be planted with a cassava variety attractive to whiteflies. Instead of Kasimbidgi green, which was used in this research, we propose the following varieties:

Kataoli	Togo
Kibandameno	Kenya
86	Ivory Coast
H58	Madagascar
CB	Zaïre

For these cultivars it is known that *B.tabaci* develops readily on them and that they are susceptable to ACMV. Unfortunately, some of these cultivars branch vigorously, which makes sampling within them difficult.

The fields should be of reasonable size, since during this study the suspicion arose that the distribution pattern of whiteflies in the field was influenced by the size of the field.

All plots in the field should be counted, because of the low numbers of *B. hancocki* that can be expected. Since the late instars are most likely to be found from leaf 20 to leaf 40 or below, it seems sensible to count all leaves of each plant, or at least the leaves from leaf 10 downwards. This would mean a lot of more work and require extra personnel.

In order to establish a culture of *B. hancocki* in our opinion a completely whitefly-proof building is required, with enough illumination to ensure adequate growth of the plants. The cages should be big enough to enable the plants to grow within them for a few months. A variety attractive to whiteflies should be used. It is recommended that temperature be controlled, since it was suspected that survival and fecundity might have been affected by high temperatures.

It would be advisable to repeat the transmission tests with more blocks and a greater number of microcages and whiteflies. Special care should be taken to select healthy plants of a sensitive variety.

Adequate whitefly material can be captured in the field, using micro cages. Greater numbers of micro cages are required however, given the long deveopmental times of the nymphs and the need for a large number of adult females.

We suggest the following points should be more carefully investigated:

- The transmission of ACMV by *B. hancocki.*
- The developmental times of *B. hancocki* nymphs (duration and number of the instars).
- The sex ratio in field populations of *B. hancocki.*
- The distribution patterns of *B. hancocki* in the field, in greater detail.
- The phenology of the *B. hancocki* populations in the field from planting until harvest.
- The influence of cassava planting date on the population dynamics of *B. hancocki.*
- The conditions required to establish and maintain a successful culture of *B. hancocki.*

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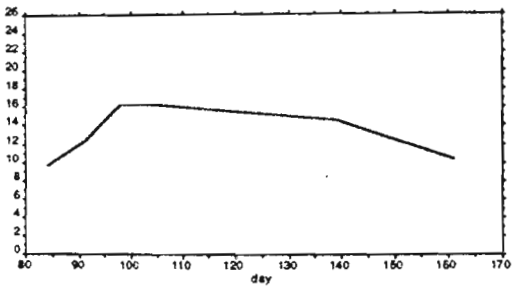
TABLES

FIGURES

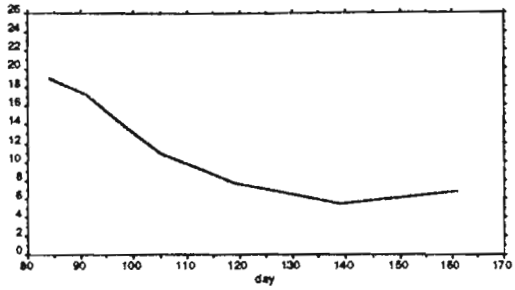
APPENDIX A.

Graphs of the change in numbers of *Bemisia hancocki* nymphs with time per plot in the first field.

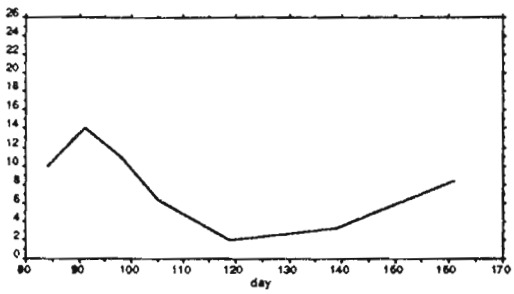
plot a1



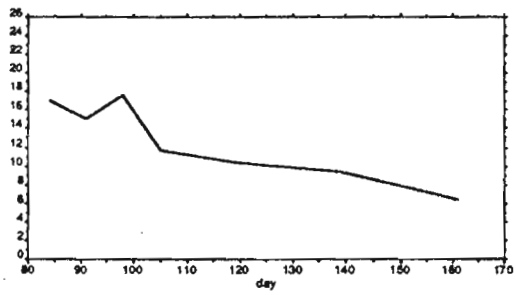
plot e1



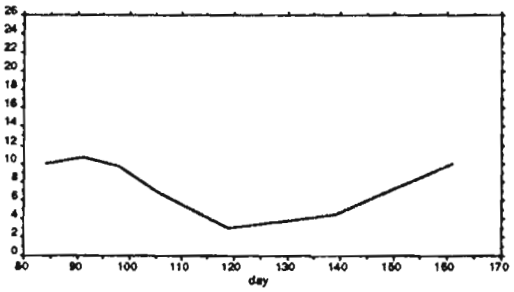
plot b1



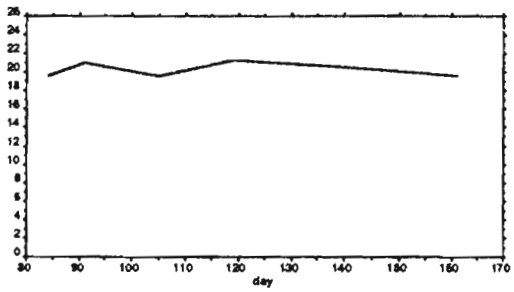
plot f1



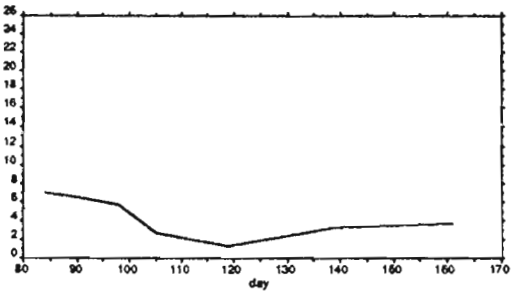
plot c1



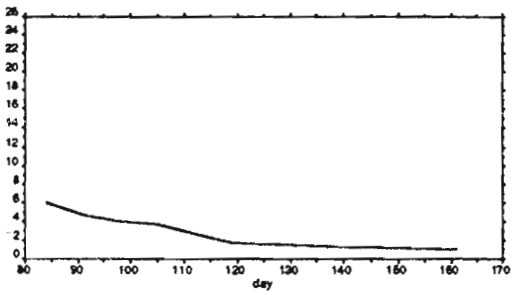
plot g1



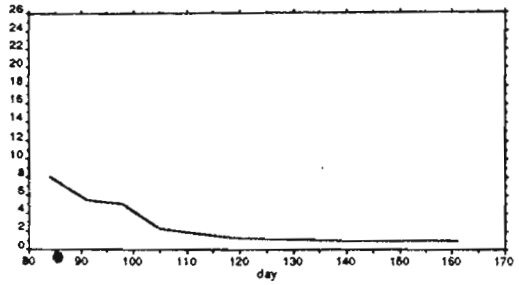
plot d1



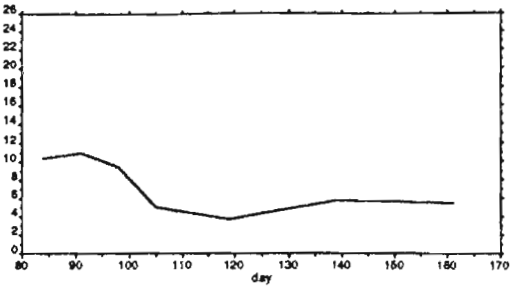
plot d2



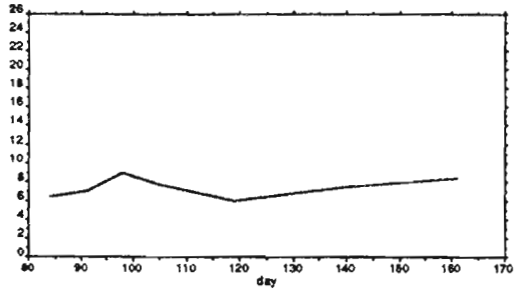
plot d3



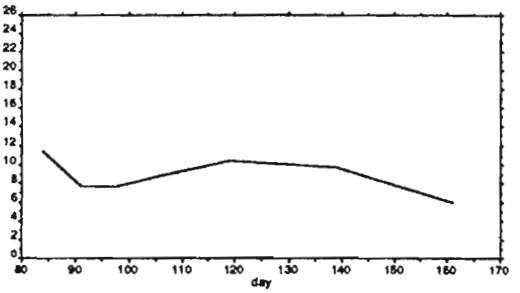
plot d6



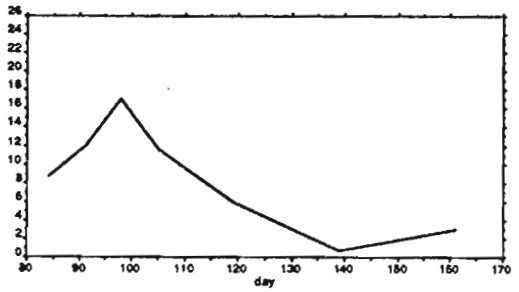
plot d4



plot d7



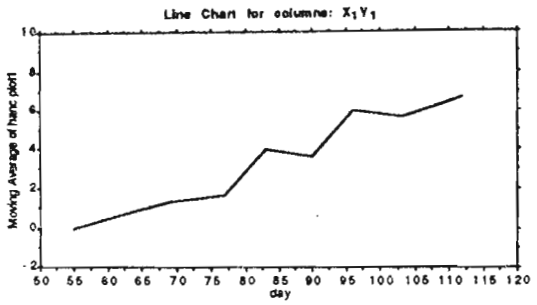
plot d5

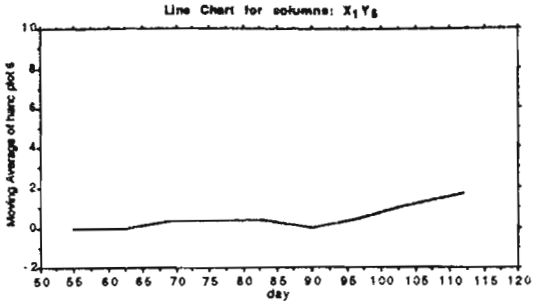


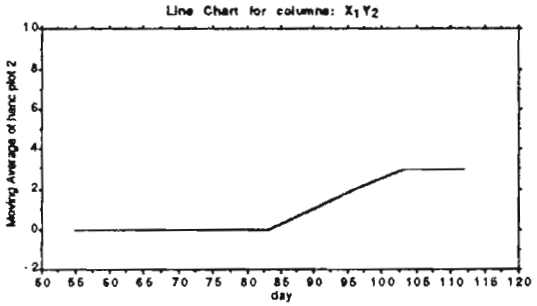
APPENDIX B.

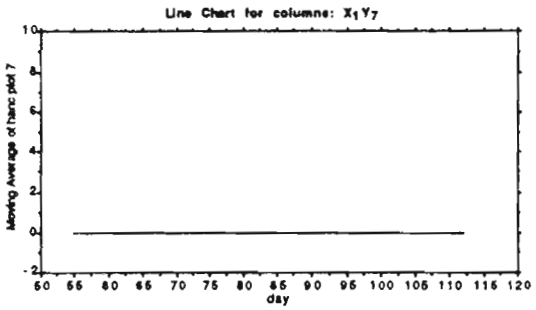
Graphs of the change in numbers of *Bemisia* nymphs with time per plot in the second field.

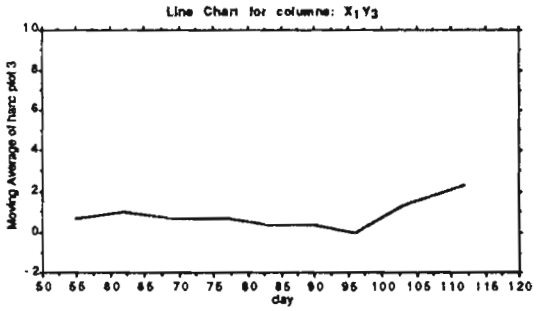
hanc1 - hanc10 = *B. hancocki* counted in plots 1 - 10
tab1 - tab10 = *B. tabaci* counted in plots 1 - 10

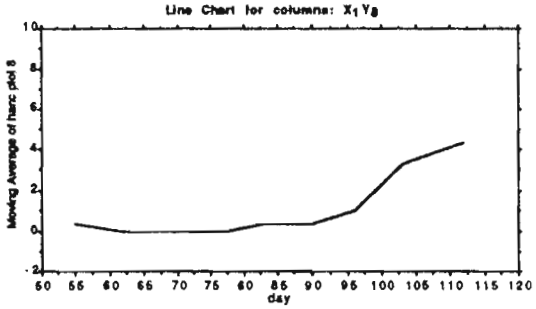

Line Chart for columns: X1 Y1
Moving Average of hanc plot1
day

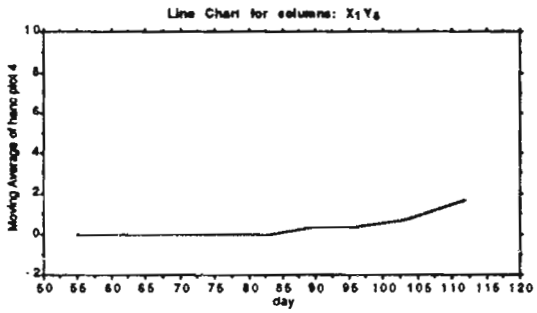

Line Chart for columns: X1 Y6
Moving Average of hanc plot 6
day

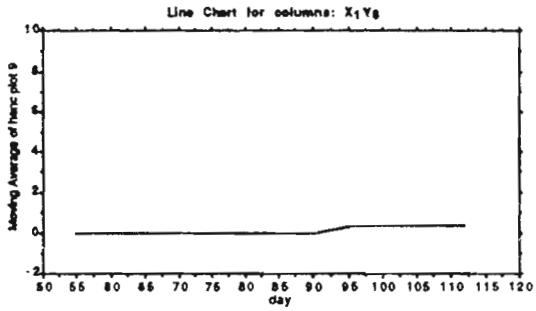

Line Chart for columns: X1 Y2
Moving Average of hanc plot 2
day

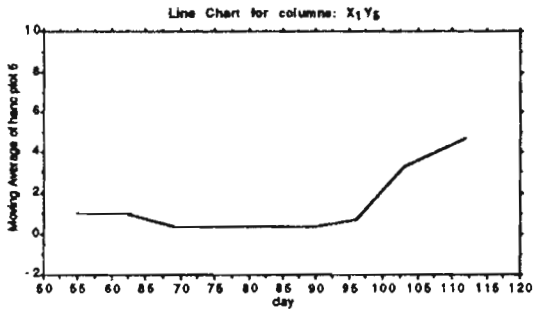

Line Chart for columns: X1 Y7
Moving Average of hanc plot 7
day

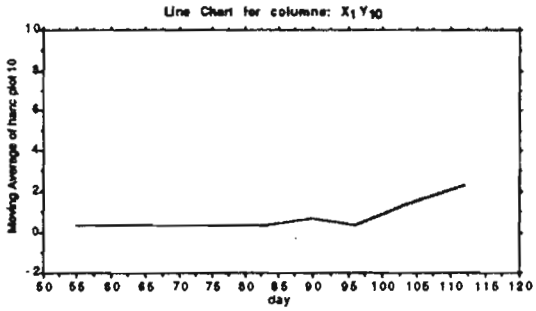

Line Chart for columns: X1 Y3
Moving Average of hanc plot 3
day

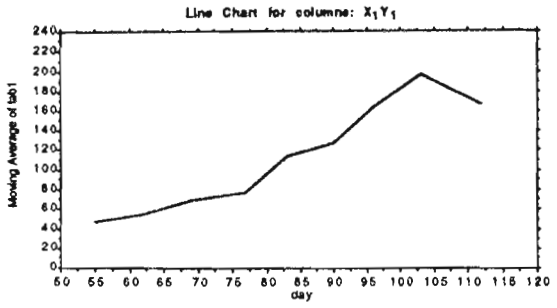

Line Chart for columns: X1 Y8
Moving Average of hanc plot 8
day

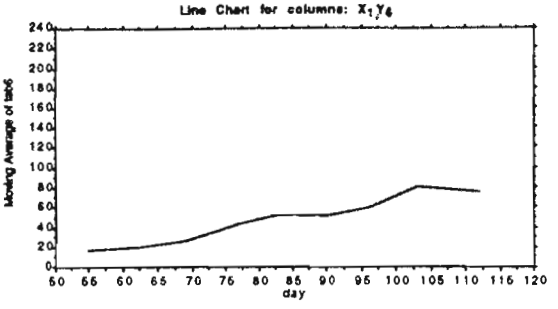

Line Chart for columns: X1 Y4
Moving Average of hanc plot 4
day

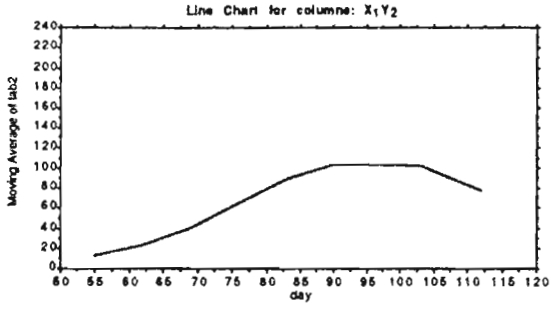

Line Chart for columns: X1 Y9
Moving Average of hanc plot 9
day

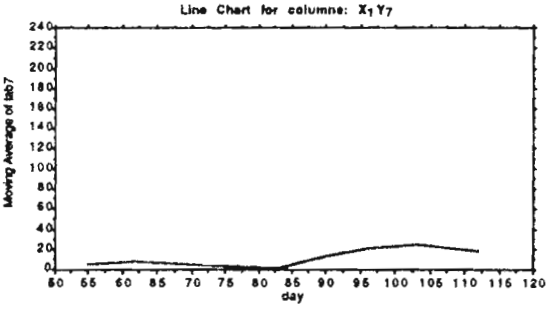

Line Chart for columns: X1 Y5
Moving Average of hanc plot 5
day

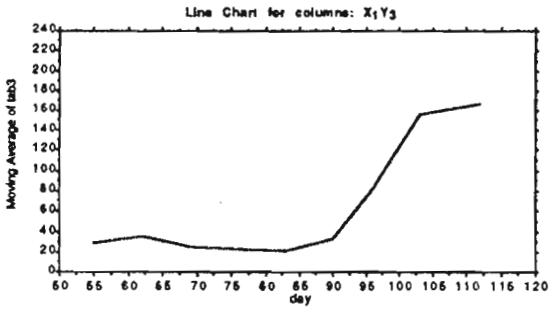

Line Chart for columns: X1 Y10
Moving Average of hanc plot 10
day

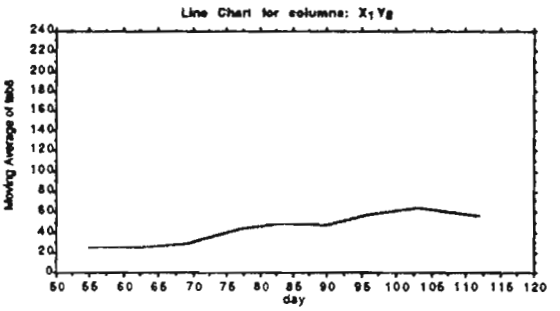

Line Chart for columns: X1Y1
Moving Average of tab1
day

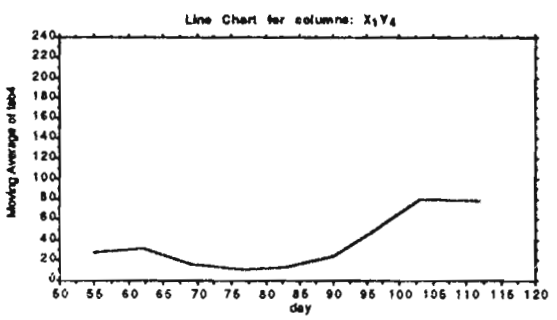

Line Chart for columns: X1Y6
Moving Average of tab6
day

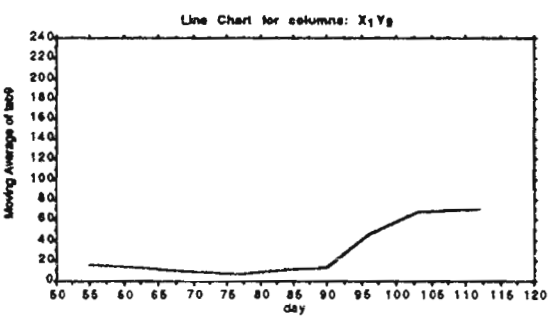

Line Chart for columns: X1Y2
Moving Average of tab2
day

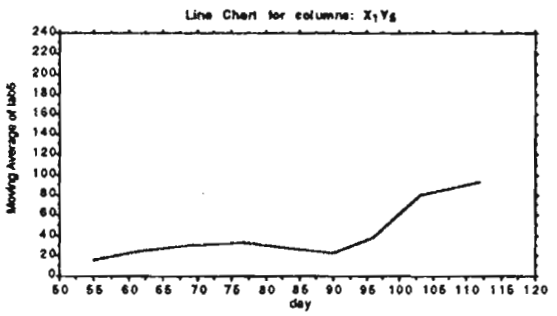

Line Chart for columns: X1Y7
Moving Average of tab7
day

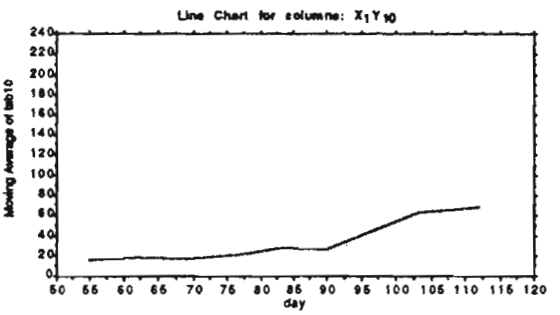

Line Chart for columns: X1Y3
Moving Average of tab3
day


Line Chart for columns: X1Y8
Moving Average of tab8
day


Line Chart for columns: X1Y4
Moving Average of tab4
day


Line Chart for columns: X1Y9
Moving Average of tab9
day

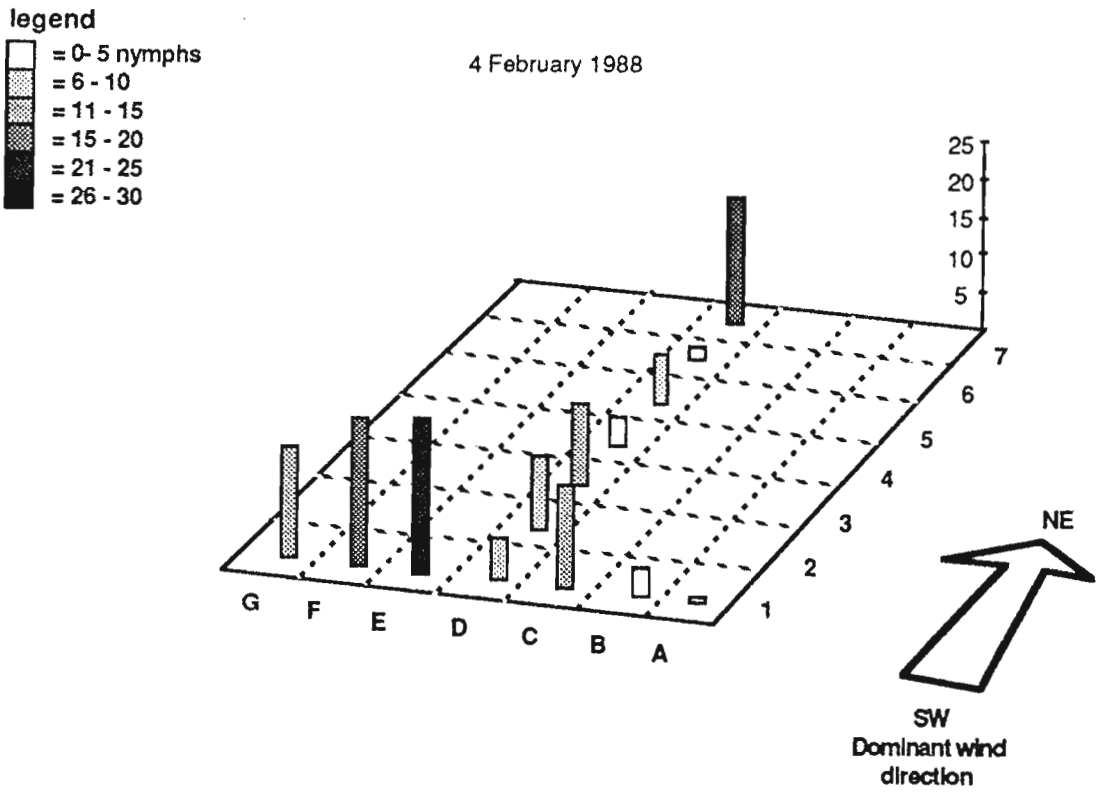

Line Chart for columns: X1Y5
Moving Average of tab5
day

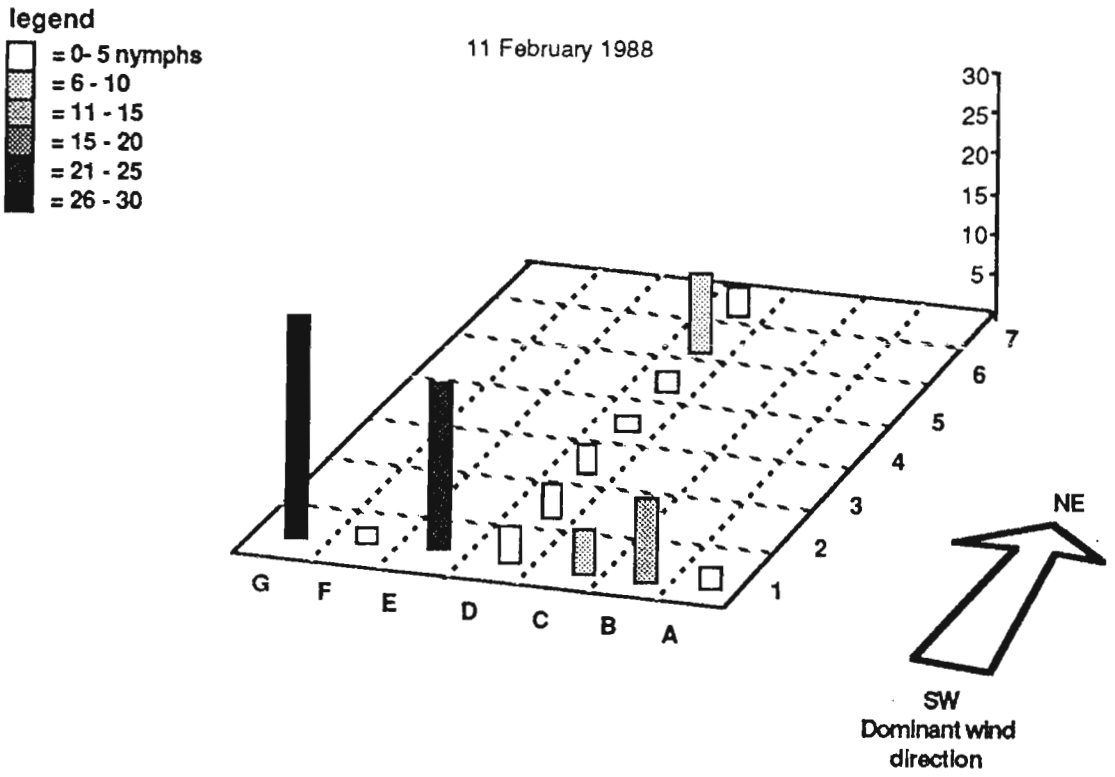

Line Chart for columns: X1Y10
Moving Average of tab10
day

APPENDIX C.

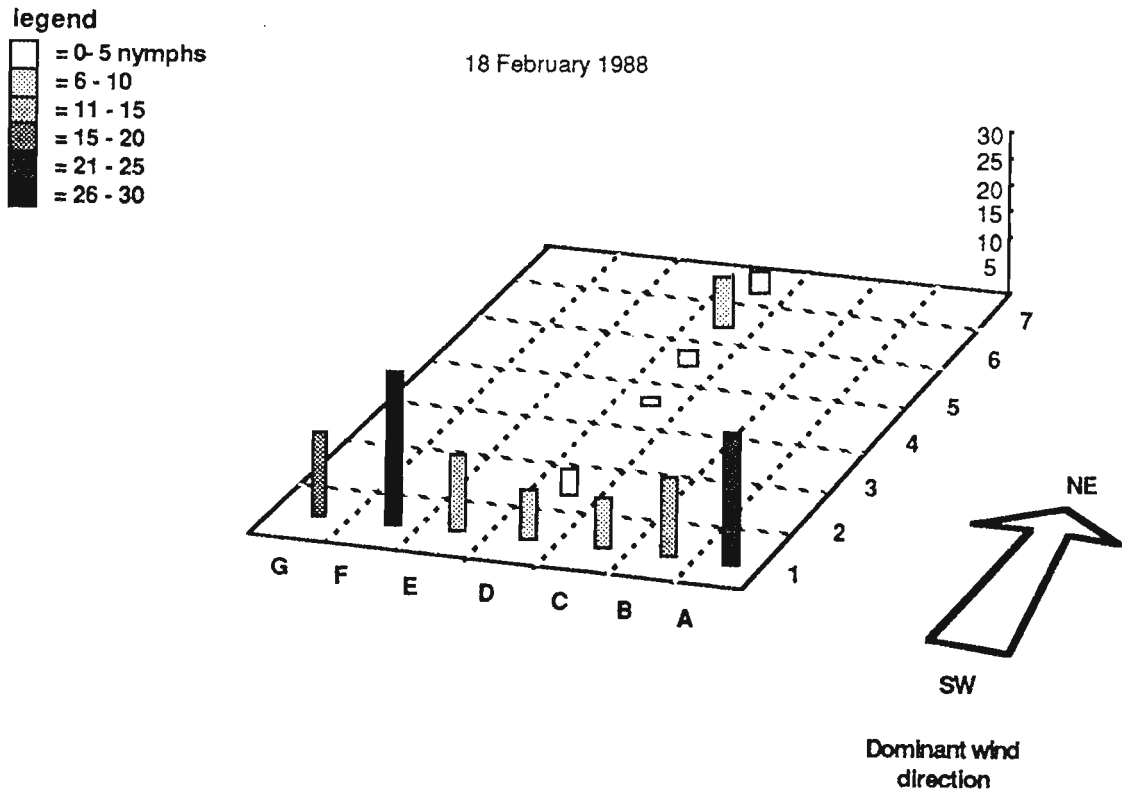
Distribution patterns of *B. hancocki* nymphs in the first field for each date.

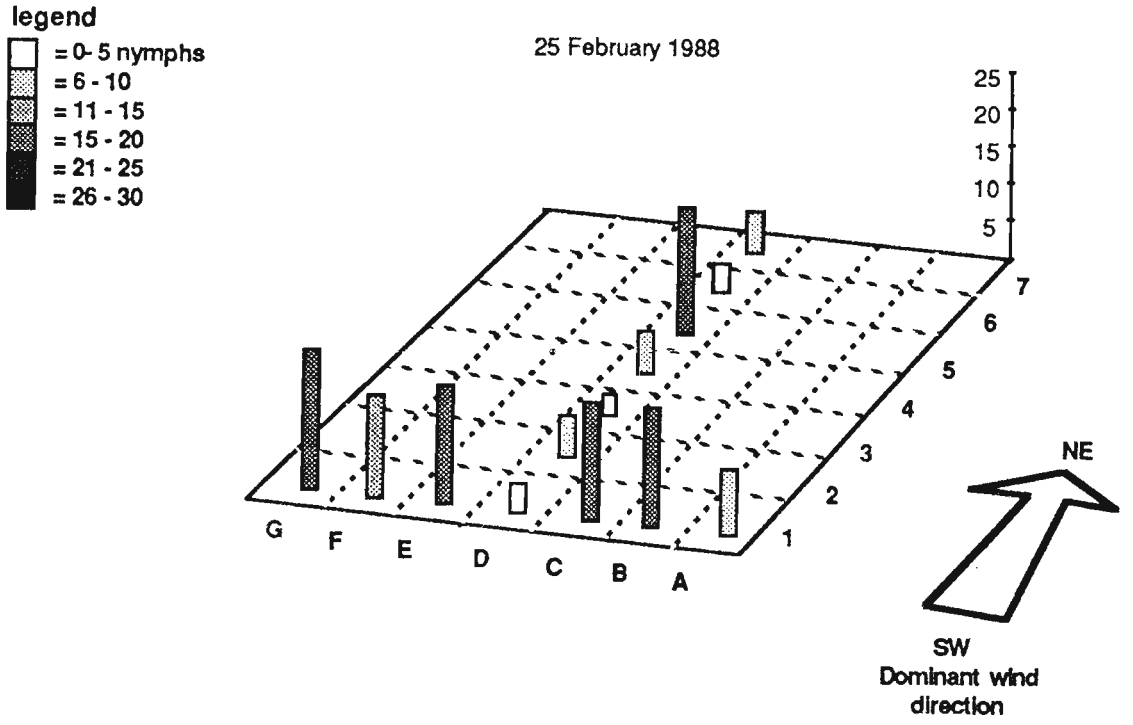
Distribution of *B. hancocki* in the field
(per plant)



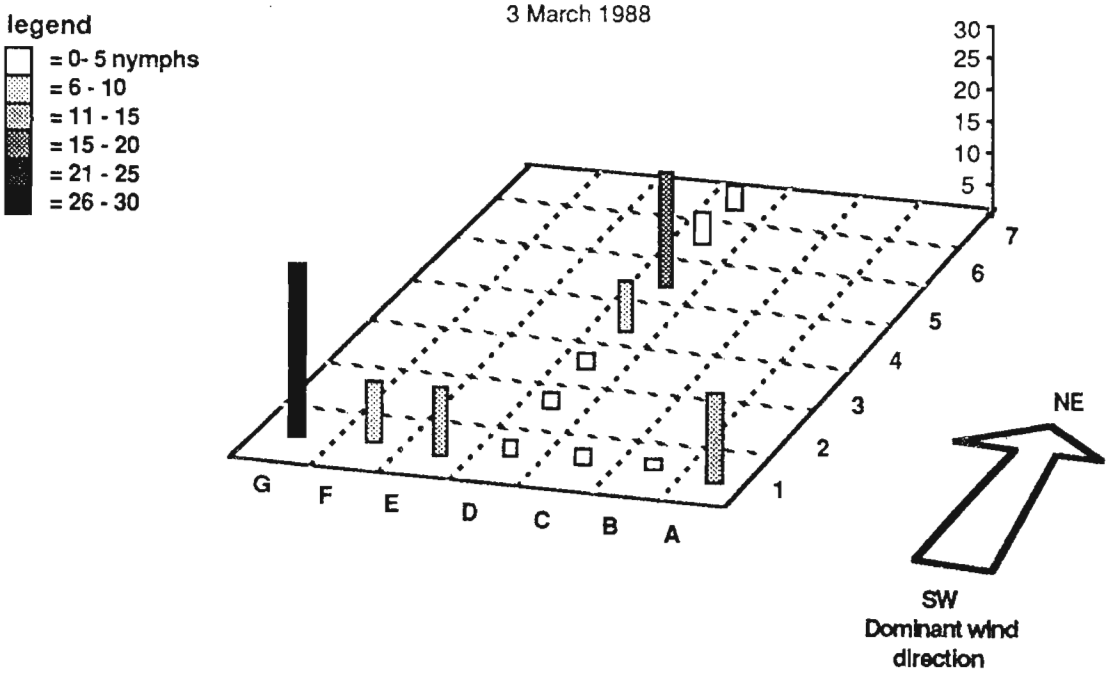


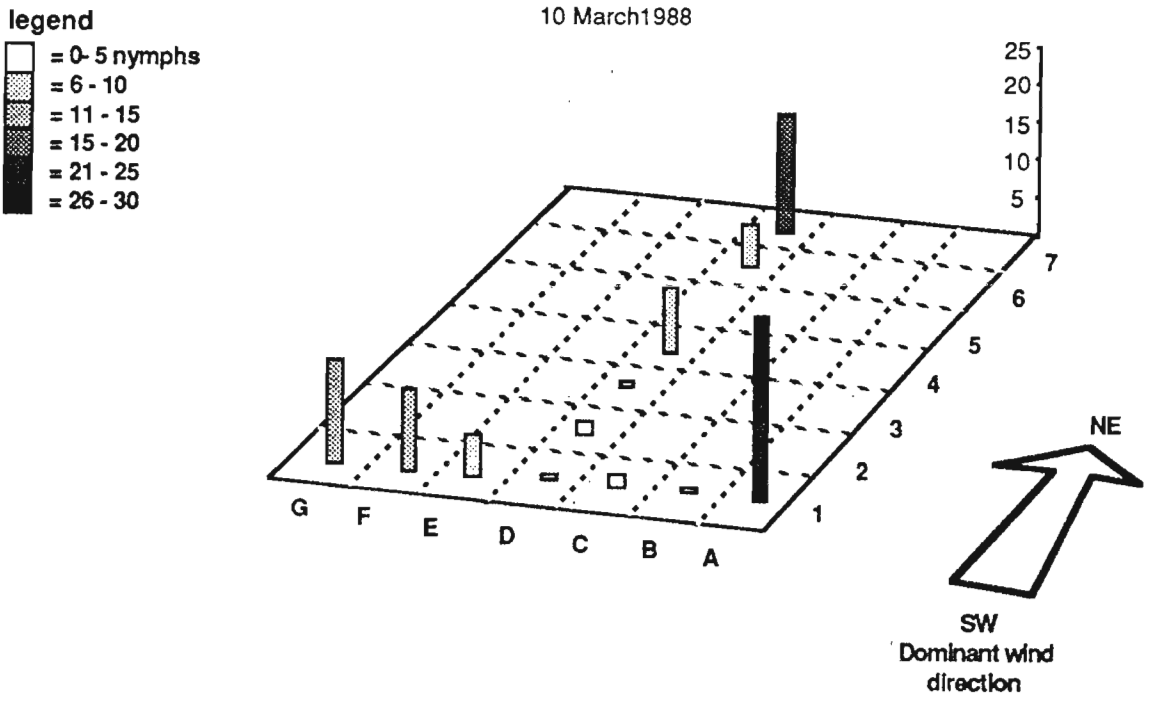
Distribution of *B. hancocki* in the field
(per plant)



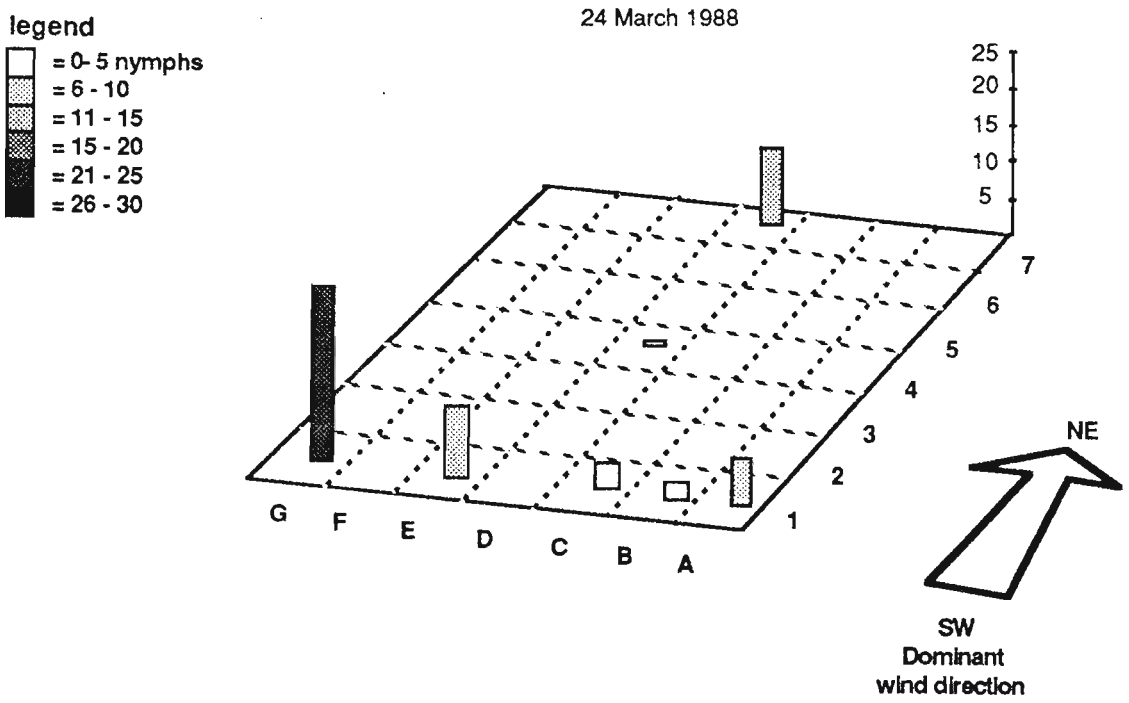


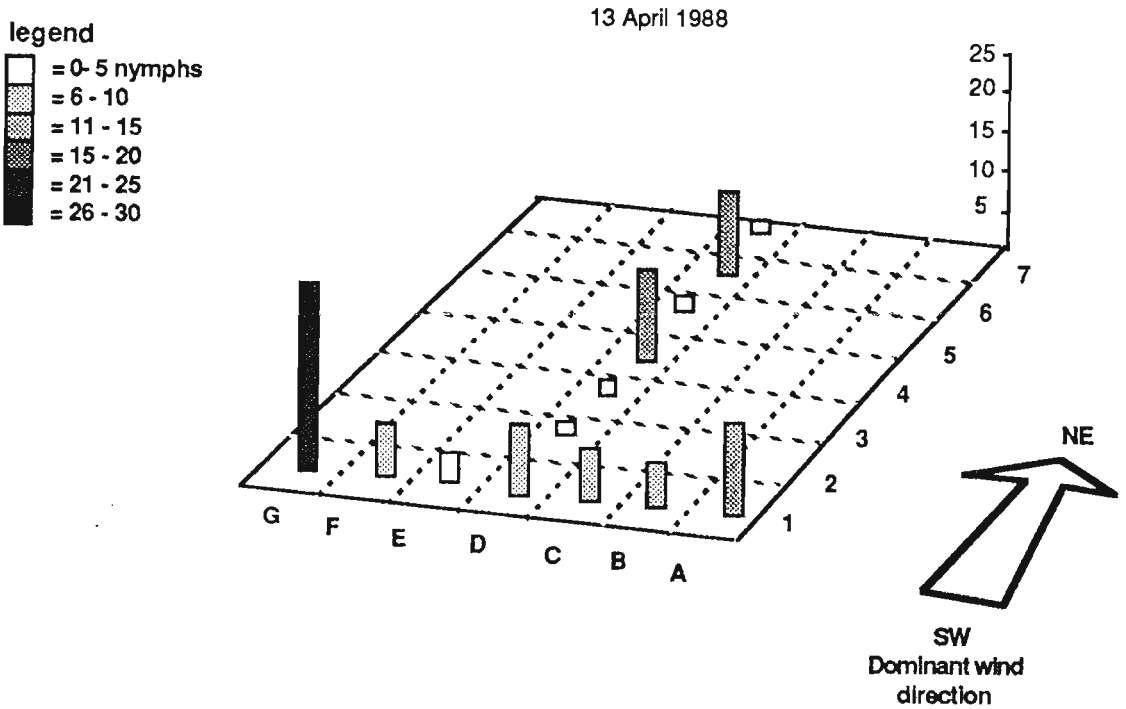
Distribution of *B. hancocki* in the field
(per plant)



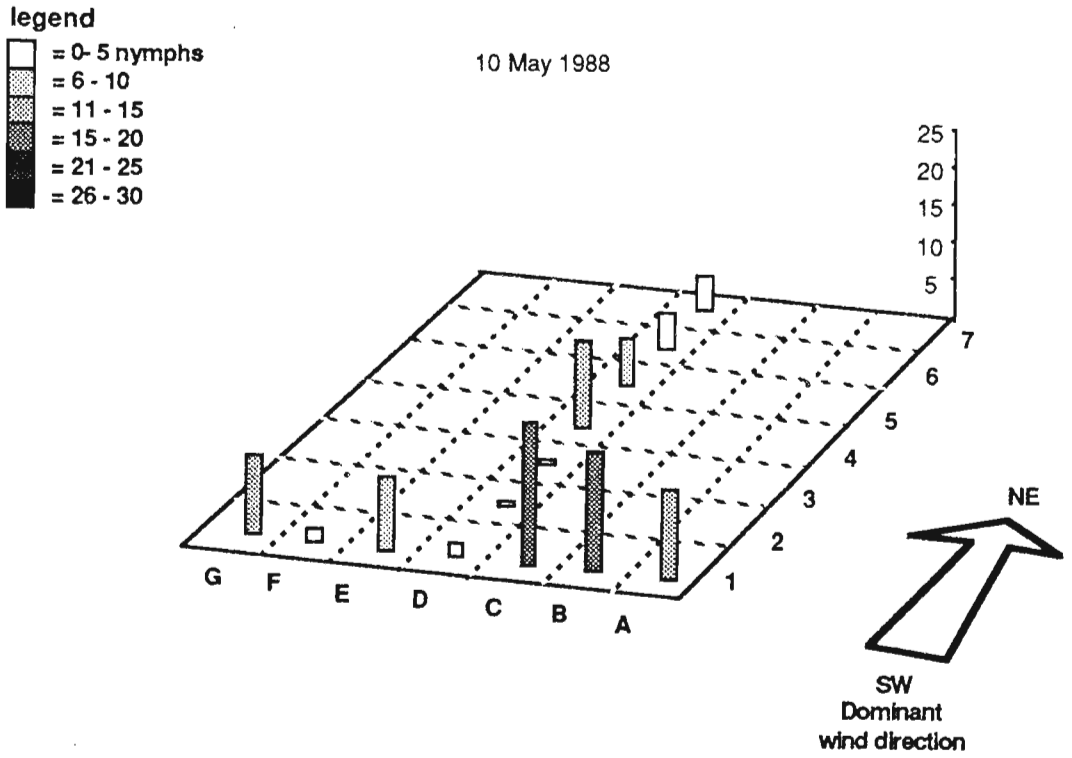


Distribution of *B. hancocki* in the field
(per plant)





Distribution of *B. hancocki* in the field
(per plant)

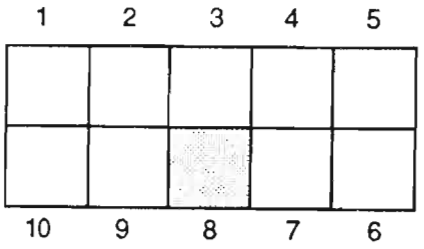


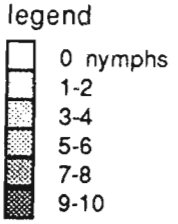
APPENDIX D

Distribution of *B. hancocki* and *B. tabaci* in the second field, per plot, per date.

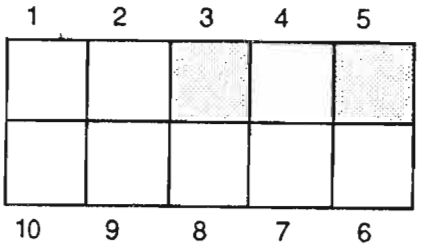
Distribution of *B. hancocki* in the field (per plant)

23 March

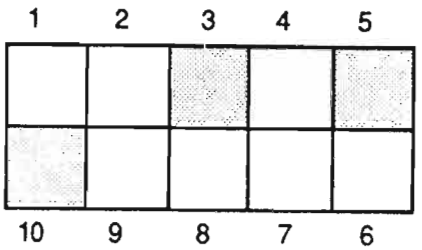




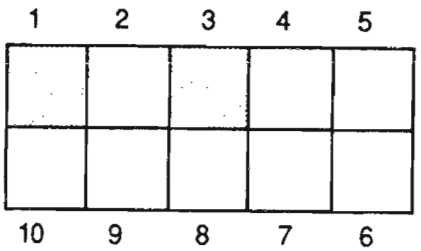
30 March



6 April

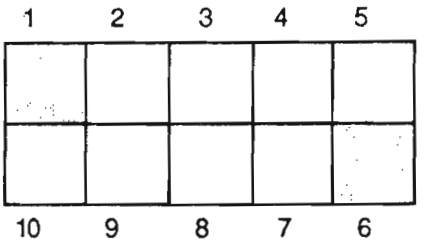


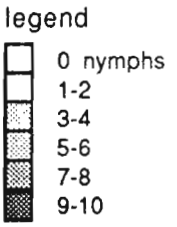
13 April



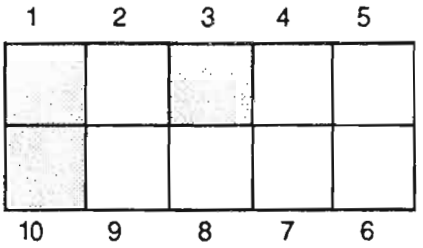
Distribution of *B. hancocki* in the field
(per plant)

20 April

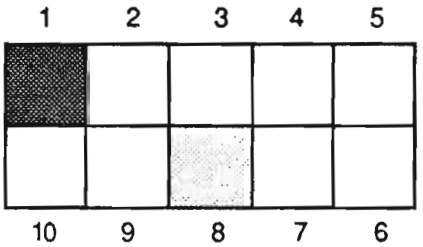




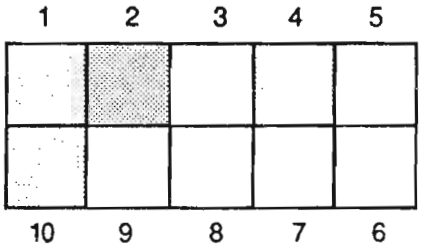
28 April



5 May

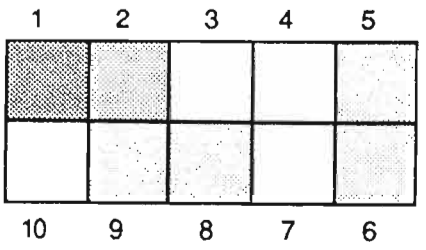


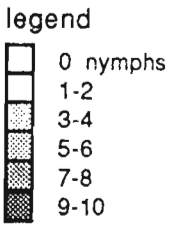
12 May



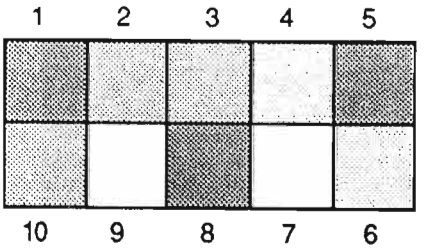
Distribution of *B. hancocki* in the field
(per plant)

18 May

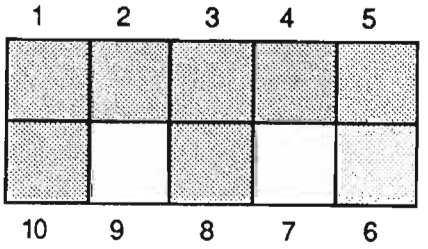




25 May

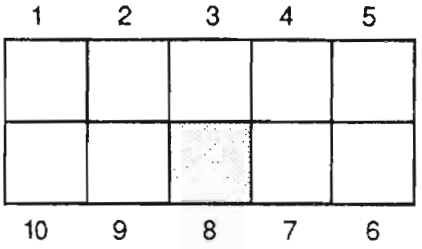


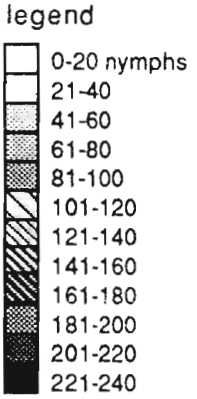
3 June



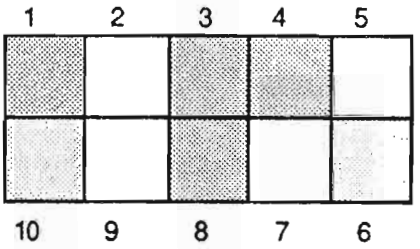
Distribution of *B. tabaci* in the field (per plant)

23 March

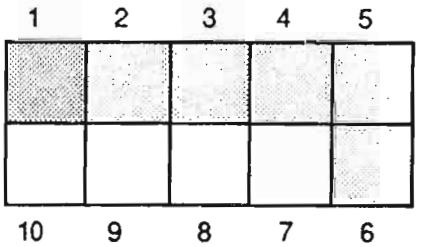




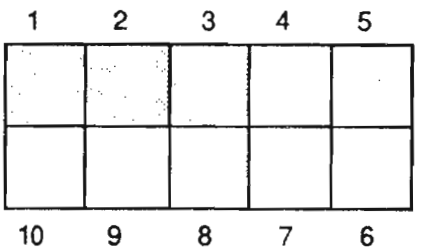
30 March



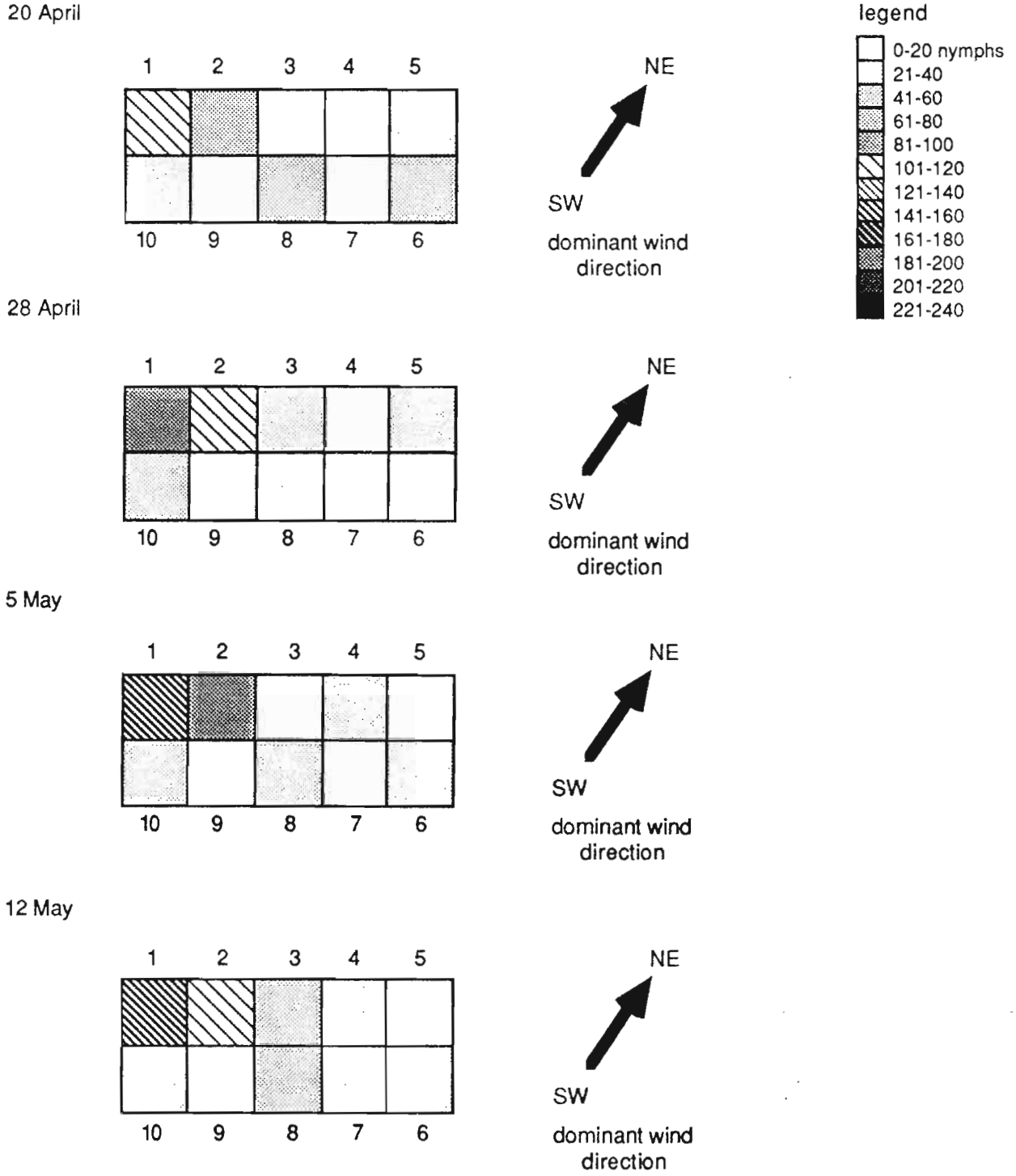
6 April



13 April

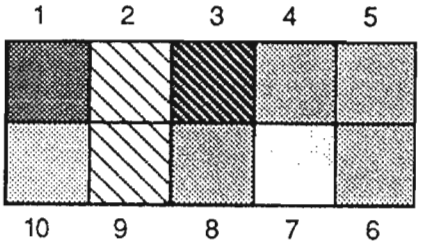


Distribution of *B. tabaci* in the field (per plant)

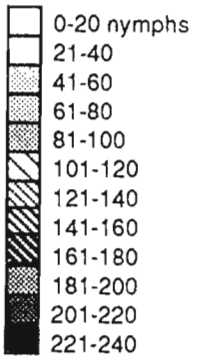


Distribution of *B. tabaci* in the field (per plant)

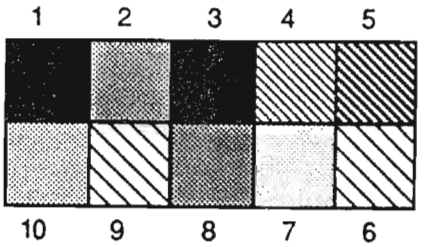
18 May



legend



25 May



3 June