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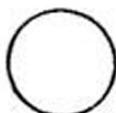
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# MECHANICAL INJURY OF WIND TO RECENTLY TRANSPLANTED CACAO SEEDLINGS AS RELATED TO THE SHADE PROBLEM

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Paulo de T. Alvim\*\*

R.M. de O. Leite\*\*\*

## ABSTRACT

Experiments in which cacao (*Theobroma cacao* L.) seedlings were submitted to different combinations of treatments involving protection and exposure to sunlight and wind have shown that the beneficial effect of shading in young cacao areas is due not only to reduced exposure to solar radiation but also to reduced air movement around the plants.

Excessive wind caused severe mechanical injury, at the pulvinus level. Visible damage occurred only 24 hours following exposure to wind. Injury progressed rapidly, leading to intensive leaf fall if the plants were maintained under non-protected conditions.

It is suggested that wind-breaks on their own can give adequate protection to cacao plantations, by providing both lateral shade and wind shelter. This practice might permit the use of a wide range of economic trees for sheltering and would probably lead to higher yield due to higher photosynthesis.

## INTRODUCTION

Several field experiments have shown that crop yields were increased

considerably when wind-breaks were used (3, 6, 7, 8, 10). Yet, little is known about the plant response to wind or the influence of the wind, alone or in

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combination with other environmental factors, on the physiology of crops. Moreover, present knowledge is almost entirely restricted to studies on the effect of wind barriers on micro-meteorological factors affecting annual crops.

There is also little information about the effects of solar radiation and wind on growth of cacao plants. Although it has been demonstrated that young cacao seedlings can thrive in the absence of shade when moisture stress is avoided by continuous watering (4), in most cacao areas it is virtually impossible to start a new plantation without shading the young plants during the first 2-3 years. This fact has been interpreted as an indication that cacao behaves as a true shade-loving species in its early stages of development. In places with very low light intensity or low potential evapotranspiration, as in some regions of Ecuador, or in areas exceptionally well protected against wind, as in the Colatina valley of Espírito Santo, Brazil, it has been possible to establish cacao plantations without shading the young plants. These, however, are exceptional cases.

After passing the juvenile stage, or when the leaf canopy is sufficiently developed to provide some self-shading, cacao growth and production are usually higher with little or no shade than when plants are shaded (1,2,5,9).

Cacao is very wind sensitive and, in areas exposed to frequent breezes,

cannot be grown without wind-breaks. Wind sheltering is a common practice, for example, in Grenada and other cacao producing islands of the West Indies, particularly in places near the coast where strong winds are frequent.

The main effect of excessive wind is to cause defoliation or premature leaf fall. In Brazil, frequent defoliation by wind occurs in Linhares, Espírito Santo, particularly when cacao is grown without or with little shade. However in the nearby valley of Colatina, which is protected by mountains, cacao has been under cultivation without shade for several years and defoliation does not occur. Annual rainfall is about the same in both places (1200-1300 mm), but in Colatina the mean wind speed is  $1\text{ m.s}^{-1}$  compared with  $4\text{ m.s}^{-1}$  in Linhares.

In some parts of Ghana and western Nigeria cacao yield is reduced by dry "harmattan" winds, which blow from the Sahara Desert between December and March. The duration and intensity of this wind vary from place to place, and are important factors in determining cacao productivity.

The present experiment is part of a larger investigation aiming to examine the effects of wind and solar radiation on cacao. Experimental wind-breaks and over-head shade were used in this work, in an attempt to evaluate the relative damages caused by each of these environmental factors on cacao seedlings.

## MATERIALS AND METHODS

Cacao (*Theobroma cacao* L.) seedlings about 4 months old were used in three experiments in the present work.

In the first experiment, the seedlings were transplanted to an unshaded area and submitted to three treatments: (a) control plants fully exposed to sunlight and without any protection against wind, (b) lateral protection against wind provided by vertical clear plastic sheets, and (c) the same as "b" but using black plastic sheets. Each wind-break measured 2.0 x 2.0m and protected nine cacao plants spaced 50cm apart. A randomized block design with four replicates was used. The leaf area per plant and the number of dead plants were measured weekly.

The same procedure was adopted in the second experiment, with the difference that two of the four replicates in each treatment received over-head shade provided by a porous black "Saram" shading cloth intercepting 60% of the incident sunlight.

As it was observed that excessive wind caused severe mechanical injury at the pulvinus level, a third experiment was carried out to evaluate the time-course of this event. Seedlings previously growing under well protected conditions were transferred to the experimental area and exposed to the various treatments applied in the sec-

ond experiment. Tissue rupture in the leaf pulvini was observed on three consecutive days.

## RESULTS AND DISCUSSION

Although severe defoliation occurred in all plants used in the first experiment during the first 2 months, presumably due to absence of over-head shade, control plants were virtually decimated, whereas plants protected by the vertically oriented plastic sheets refoliated well (Figure 1). Plants surrounded by black and clear plastic sheets were equally defoliated in the first two months but those protected by black sheets refoliated better, probably because of the development of higher air temperatures around the plants.

The second experiment investigated whether or not the defoliation that occurred in the first experiment was due to the absence of over-head shade. The results (Figure 2) showed that over-head shade was effective in preventing leaf fall only when associated with the use of wind-breaks. Wind sheltering, on the other hand, furnished sufficient protection against defoliation even in the absence of over-head shade. Thus, the plants protected against wind and direct solar radiation did not differ significantly from those exposed to sunlight but surrounded by wind-breaks. In fact, no difference was observed when black plastic sheets were used as

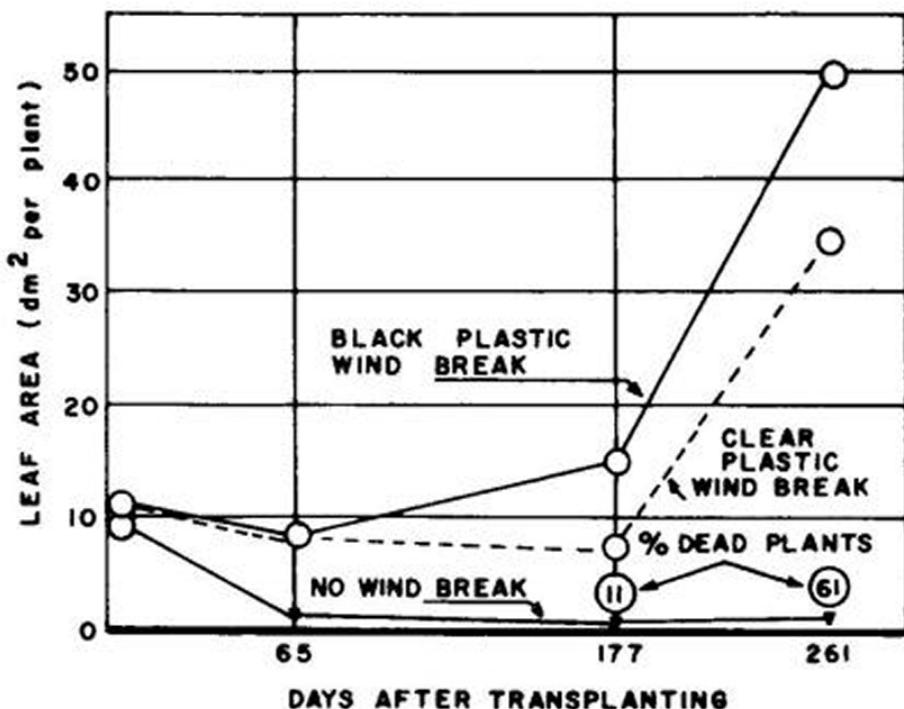


Figure 1 – Effect of black and clear plastic "wind breaks" on leaf area and percentage of dead seedlings, following transplanting to an unshaded site.

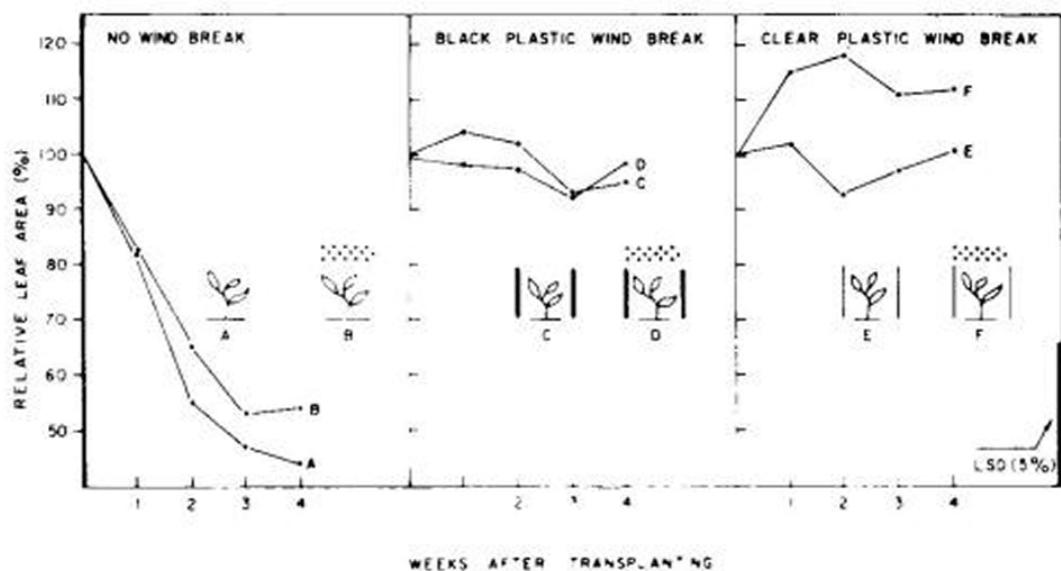


Figure 2 – Interacting effects of black and clear plastic wind breaks and over-head shade on leaf area of recently transplanted cacao seedlings. A – plants fully exposed to sunlight and without wind protection; B – over-head shade, no wind break; C – no shade, black plastic wind break; D – over-head shade, black plastic wind break; E – no shade, clear plastic wind break; F – over-head shade, clear plastic wind break.

wind-breaks. Though not significant, the differences found between wind-protected shaded and unshaded plants (F and E), as well as those observed between non-protected shaded and unshaded plants (B and A) may well be due to the possibility that the shading "Saram" cloth also afforded some wind sheltering.

The high defoliation rate that occurred in wind sheltered plants in the first experiment was probably due to less favorable climatic conditions following transplant.

As previously mentioned, the pulvini situated at the base of the leaf blades

suffered severe mechanical injury when the plants were exposed to excessive wind. This tissue, formed by a mass of thin-walled cells, showed visible rupture in its outer layers only 24 hours following exposure to wind. The injury progressed rapidly, attaining the vascular strands within a few days and finally causing leaf fall if the plants were maintained under non-protected conditions.

Figure 3 shows the percentage of damaged pulvini in the six environmental conditions employed in the third experiment. Control plants fully exposed to sunlight and without any

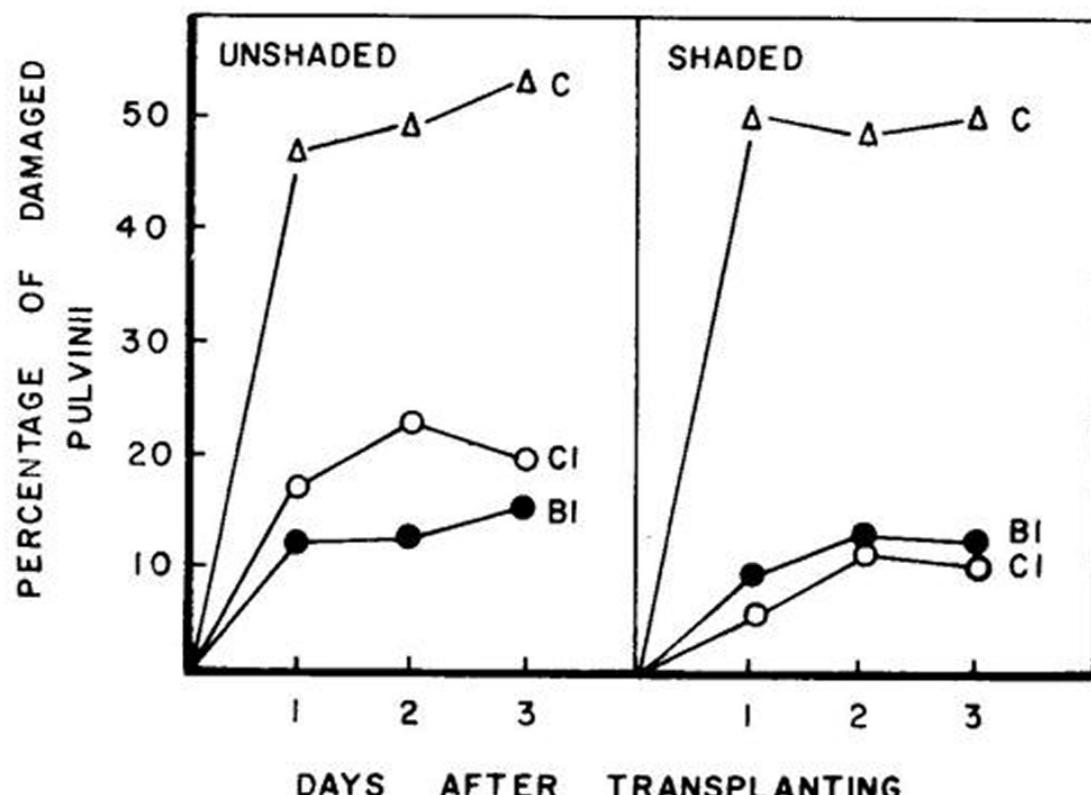


Figure 3 - Mechanical damages of wind to the pulvini of cacao seedlings following transplanting to shaded and unshaded conditions (C = control plants with no wind break, CI = clear wind break, and BI = Black plastic wind break).

protection against wind had nearly 50% of their pulvini showing the initial epidermic rupture on the next day following transplant. When clear plastic sheets were used as wind-breaks, a significant difference in damage was found between plants growing with and without over-head shade, the former being more effectively protected, presumably due to an excessive water loss in the plants which were more exposed to sunlight.

This study indicates that the beneficial effect of shading in young cacao areas is due not only to reduced exposure to solar radiation but prima-

rily to reduced air movement around the plants.

The results found in the present work suggest that wind-breaks on their own can give adequate protection to cacao plantations, by providing both lateral shade and wind shelter.

Through the use of wind-breaks it is foreseen that the cacao plants would grow faster and yield more due to higher photosynthesis. This might permit the use of economic trees for sheltering which had previously not been considered for overhead shading because of the undesirable shape of their canopy.

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## RESUMO

### **Injúria Mecânica do Vento Sobre Mudas de Cacau Recém-Transplantadas com Relação ao Problema do Sombreamento**

Experimentos em que plântulas de cacau (*Theobroma cacao* L.) foram submetidas a diferentes combinações de tratamentos, envolvendo proteção e exposição à luz e ao vento, demonstraram que o efeito benéfico do sombreamento de novas plantações de cacau deve-se não somente à exposição a baixas intensidades luminosas, mas também à redução da turbulência do ar em torno das plantas.

Ventos excessivos ocasionaram severa injúria mecânica na região do pulvinulus. Danos visíveis ocorreram apenas 24 horas após exposição ao vento. Os efeitos deletérios progrediram rapidamente, provocando intensa queda de folhas quando as plantas eram mantidas desprotegidas.

Sugere-se que uma proteção adequada pode ser oferecida a cacauais por barreiras periféricas que forneçam sombreamento lateral e redução na velocidade dos ventos, em substituição ao sombreamento de topo convencionalmente uti-

lizado. Tal prática permitiria selecionar maior variedade de espécies econômicas para a proteção ambiental de cacauais e provavelmente conduziria a produtividades mais altas, em decorrência de maior fotossíntese.



# SCREENING OF CACAO CULTIVARS FOR RESISTANCE TO *Phytophthora palmivora* IN THE COLLECTION AT CATIE, COSTA RICA

Jeremy S. Lawrence \*

## ABSTRACT

Screening of the CATIE cacao collection for resistance to *Phytophthora palmivora* was initiated, using point-inoculation with zoospore suspension of unwounded attached pods to evaluate cultivar response. Of the 51 cultivars tested, 9 showed a promising degree of resistance; EET 59, EET 376, Pound 7, UF 713, UF 715, Scavina 6, Scavina 12, Catongo, Diamantes 800.

## INTRODUCTION

Losses in cacao due to *Phytophthora palmivora* (Butler) Butler (*Phytophthora* pod rot disease) can be reduced by cultural methods and by the use of fungicides. However, these practices, especially fungicide applications, are often costly in relation to the quantity of cacao beans saved and many growers regard them as economically unfeasible, particularly when

cacao prices are low. The use of resistant varieties is often the most effective and economic means of controlling plant diseases, and the replacement of susceptible cacao trees by material showing durable race non-specific resistance to *P. palmivora* would provide the ideal, long-term solution to combatting the disease.

Unfortunately, amongst those cacao cultivars in the world whose

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reaction to *P. palmivora* is known, most exhibit varying degrees of susceptibility to *Phytophthora* pod rot and so far no immune cultivars have been found, although a number in different areas have been reported to show relatively low levels of susceptibility (9, 10, 13). Consequently, the search for resistant parent material remains a basic requirement in breeding for resistance to *P. palmivora*.

The larger cacao collections of the world may provide valuable sources of resistance since the reaction to *P. palmivora* infection is still unknown for many of their accessions. For this reason, systematic screening of the CATIE collection was initiated with the purpose of identifying promising resistant cultivars which could be of use in breeding programmes. This report presents the results obtained up to July 1977.

## MATERIALS AND METHODS

A previous study in Costa Rica (4) showed that the most reliable and consistent method for screening fruiting cultivars was point-inoculation with standard zoospore suspension of attached pods without wounding, the method adopted here. Pods of known age, 1 month from maturity, were used and whenever possible only one or two were selected from each tree. Ten replicate pods per cultivar were inoculated and when sufficient

pods were available each cultivar was tested at least twice at different times. Both percentage successful infection and average lesion diameters were recorded, the latter when lesion sizes on pods of UF 677, a standard, highly susceptible cultivar included in all tests, attained an average diameter of 6 cm. When cultivars were tested twice or more, percentage infection and average lesion diameters were calculated from the sum of all inoculated pods.

The same single-sporangium *P. palmivora* isolate (morphological form 1, compatibility type A2), selected on the basis of pathogenicity on pods, was used in all tests. To maintain infective potential, the isolate was passed through pod tissue every 1–2 months. Zoospore suspensions at a concentration of  $2 \times 10^5$  spores/ml were prepared as described by Lawrence (4).

All cultivars examined were cloned material and, whenever possible, higher yielding cultivars or those suspected from field observations of possessing better resistance to *P. palmivora* were tested. All tests were carried out between April and December each year, a period which encompassed peaks in *P. palmivora* incidence and which coincided with the times of higher pod production and greatest rainfall.

## RESULTS AND DISCUSSION

Of the 51 cultivars examined, nine demonstrated a promising degree of

resistance; EET 59, EET 376, Pound 7, UF 713, Scavina 6, Scavina 12, Catongo, Diamantes 800 and UF 715 (Table 1). Catongo was obtained as cuttings from one of the original selections in Bahia, Brazil, and Diamantes 800 is a selection from the Diamantes Experimental Station in Costa Rica.

With these nine cultivars, as well as CC 42, even when percentage infection was high, lesions developed very slowly, often ceasing expansion altogether 2-3 days after visible symp-

toms first appeared, thus, indicating resistance to post-penetration development by *P. palmivora*. On the basis of percentage infection, EET 376, Scavina 6, Scavina 12, and especially EET 338 appeared to be more resistant to epidermal and epicarp penetration by *P. palmivora*. However, when lesions formed on EET 338, they developed as rapidly as on the most susceptible cultivars. Therefore, only EET 376, Scavina 6 and Scavina 12 showed resistance to both initial penetration and post-penetration growth by *P. palmivora*.

Table 1 - Responses of cacao cultivars to point-inoculation with *P. palmivora* zoospore suspension of unwounded attached pods.

Cultivar	% successful infection*	Av lesion diam (cm)§	Cultivar	% successful infection	Av lesion diam (cm)
EET 59	100	0.4	IMC 67 <sup>+</sup>	90	3.5
EET 376	50	0.5	EET 397 <sup>+</sup>	100	3.5
Pound 7	70	0.6	UF 12	90	3.6
UF 713	90	0.6	UF 668	85	3.8
Scavina 6	40	0.65	UF 168 <sup>+</sup>	95	4.3
Scavina 12	55	0.7	UF 10 <sup>+</sup>	100	4.6
Catongo	95	0.7	Pound 12	80	5.0
Diamantes 800	80	0.75	P 16 <sup>+</sup>	100	5.1
UF 715	70	0.8	UF 29	100	5.1
UF 704	75	1.0	UF 708 <sup>+</sup>	95	5.2
CC 42	85	1.0	UF 122	95	5.2
CC 38 <sup>+</sup>	90	1.2	UF 650	100	5.4
UF 613	95	1.2	EET 75 <sup>+</sup>	100	5.4
SIC 433 <sup>+</sup>	100	1.5	UF 676	100	5.5
EET 156 <sup>+</sup>	70	1.9	UF 667	95	5.6
UF 36 <sup>+</sup>	100	2.5	UF 221	100	5.7
UF 601	95	2.6	UF 677	90	6.0
UF 707 <sup>+</sup>	85	2.9	R 56 <sup>+</sup>	100	6.1
CAS 3 <sup>+</sup>	100	2.9	R 13 <sup>+</sup>	100	6.1
UF 296	95	3.0	R 10	90	6.2
CC 41	100	3.0	GS 36 <sup>+</sup>	100	6.2
UF 93 <sup>+</sup>	100	3.1	R 52 <sup>+</sup>	95	6.3
CC 45 <sup>+</sup>	90	3.2	EET 338	35	6.5
UF 701 <sup>+</sup>	90	3.2	SGU 71 <sup>+</sup>	100	7.1
CC 10	100	3.3	EET 353 <sup>+</sup>	100	8.0
UF 654	95	3.4			

\* 2 opposite, lateral inoculum points on each of 10 replicate pods per test.

§ measured when av lesion diam on the "standard" susceptible cultivar UF 677, included in each test, attained 6cm.

+ cultivar tested only once.

With respect to pod losses under natural conditions, resistance to epidermal and epicarp penetration alone may be a less important character than resistance to post-penetration, as exemplified by EET 59 and EET 338. Although infection may be low with EET 338, whenever it does occur the pods rot quickly. With EET 59, proportionately many more pods may be infected but probably fewer become rotted before harvest. The net result, therefore, is that, other things being equal, EET 338 probably suffers greater pod losses than EET 59. Field observations tended to support this hypothesis but pod or bean loss data were not available to confirm it.

Nevertheless, post-penetration resistance which retards the rate of pod tissue colonization by *P. palmivora* and which, consequently, reduces the risk of bean infection before harvest, is a more desirable form of resistance than that to initial penetration alone. Whenever this latter type of resistance is overcome by the pathogen, there is no further resistance to growth within pericarp tissues and rapid rotting of the pod ensues.

Responses to *P. palmivora* infection of a number of the cultivars have been reported previously, from Costa Rica and elsewhere, evaluation being either by natural infection in the field or by artificial inoculation methods (1, 2, 3, 5, 6, 7, 8, 11, 12, 14, 15, 16, 17). With most of these cultivars there is good agreement in their responses

to *P. palmivora* between previous reports and the present results. However, some notable differences exist. CC 41 and UF 29 were reported as resistant under field conditions in the Atlantic Zone of Costa Rica (1, 15) but these two cultivars show disease escape in that area (1) and their intrinsic susceptibility has been established (1,4).

Measuring development of *P. palmivora* mycelium in liquid media incorporating pod-husk tissue, Orellana (7) in Costa Rica graded UF 12 as resistant, in contrast to the moderately susceptible reaction reported here. However, methods utilizing pod-tissue extracts fail to take into account possible physical and chemical resistance mechanisms imparted by or present in intact epidermal and pericarp tissues, so that differences in cultivar response between these methods and inoculations of intact pods could be expected.

Using a stem-inoculation method, Zentmyer (17) reported that UF 715 was highly susceptible. Since this cultivar was tested with a Costa Rican A2 isolate from cacao, the discrepancy with the present results is hard to explain. Stem inoculations and inoculations of attached pods have been shown to be similarly reliable and consistent as methods for assessing resistance to *P. palmivora* (4), so such drastic differences in cultivar response between the two methods would not be expected. So far, all *P. pal-*

*mivora* isolates from cacao in Costa Rica have been identified as morphological form 1, so it seems unlikely that the discrepancy was due to the use of different morphological forms.

Of the nine most resistant cultivars mentioned here, responses to *P. palmivora* of only four others in addition to UF 715 have been reported previously, all being classified as resistant; Pound 7 (8), Scavina 6 (3, 8, 11, 12, 16, 17), Scavina 12 (11, 16) and Catongo (5, 6, 12, 14, 17). In future screening of the CATIE collection it is recommended that the following potentially promising cultivars be

examined: EET 19, 48, 62 and 64; CC 9, 17, 34, 48, 69, 107, 124, 137, 152 and 178; CAS 1 and 2; PA 169; CATIE 1000.

It should be emphasized that the reaction to *P. palmivora* infection of all cultivars, including those reported here, only applies to those areas where they were tested and to those strains of *P. palmivora* with which they were infected. Cacao types reported as more resistant may well be much more susceptible in other cacao-growing regions where different environmental conditions might prevail and where other morphological forms and races of *P. palmivora* might be present.

#### ACKNOWLEDGMENTS

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## RESUMO

### Seleção para Resistência a *Phytophthora palmivora* em Cultivares de Cacau da Coleção de CATIE, Costa Rica

Foi iniciada a seleção para resistência a *P. palmivora*, na coleção de cacau de CATIE, usando-se inoculação pontual com suspensão de zoósporos em frutos não destacados e sem ferimentos para avaliação da resposta dos cultivares. Dos 51 cultivares testados, nove apresentaram grau de resistência promissor, a saber: EET 59, EET 376, Pound 7, UF 713, UF 715, Scavina 6, Scavina 12, Catongo, Diamantes 800.



# WATER-HOLDING PLANTS (PHYTOTELMATA) AS LARVAL HABITATS FOR CERATOPOGONID POLLINATORS OF CACAO IN BAHIA, BRAZIL

Durland Fish \*

Saulo de J. Soria \*\*

## ABSTRACT

Ten samples of three types of water-holding plants (Phytotelmata) were examined at the end of a dry period in the cacao growing area of Bahia, Brazil for larvae of Ceratopogonidae (Diptera), potential pollinators of *Theobroma cacao* L. A wild flower *Calathea* sp. (Marantaceae) contained an average of 22.4 *Culicoides* n. sp. in the inter-bract liquid of its inflorescence, *Musa* (AAB Group) "banana prata" (Musaceae) contained an average of 2.4 *Forcipomyia (Warmkeia)* sp. within its water-filled leaf-axils, and an epiphytic tank bromeliad *Vriesea procera* (Bromeliaceae) contained an average of 3.0 *F. (Phytohelea) caribbeana* also within water-filled leaf-axils. Ceratopogonids in general were dominant in the *Calathea* community, co-dominant in banana, and third in abundance in bromeliads. Phytotelmata associated with cultivated cacao are indicated to be favorable habitats for specific ceratopogonid species. Increasing the diversity and abundance of water-holding plants may therefore increase the chances of more insect species participating in the pollination process, particularly during dry weather. However, the potential of bromeliads as sources of medically important insects, such as malaria mosquitoes, must also be considered in any attempts to manipulate the phytotelm flora in cacao plantations.

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## INTRODUCTION

Successful pollination of cacao flowers is an important limiting factor in the production of cacao in many areas of the tropics (7, 14). The inadequacy of the natural pollination process in cultivated cacao is evidenced by a wide geographic variation in pollination rates (7, 14) and a marked increase in pollination success in areas where mechanical methods have been experimentally employed (5, 14, 15).

The fact that *Theobroma cacao* is dependent upon insect pollination has been well established. In Ghana, pollination has been attributed to psyllids, hymenopterans, cecidomyiids, and other small insects including three genera and eight species of ceratopogonids (3, 4). In Costa Rica, Soria (14) reported ceratopogonids of four subgenera of *Forcipomyia* as well as thrips to be important pollinators, and in Trinidad, Macfie (6) reported ceratopogonids of both *Forcipomyia* and *Lasiohelea* to be important. Soria and Wirth (16) found the subgenus *Euprojoannisia* of *Forcipomyia* to be important pollinators of cacao in Bahia, Brazil, and there is evidence that other genera of ceratopogonids are involved (19). Although the total insect fauna participating in the pollination of cultivated cacao is not precisely known, ceratopogonids, in particular the genus *Forcipomyia*, seem to be important in most areas.

Recently, much emphasis has been placed on locating the natural larval

habitats of ceratopogonids in cacao plantations (3, 17, 19, 20). Opened cacao pods, rotting cacao fruits, leaf litter, rotting banana stems, and other decomposing organic debris have proved to be sources of a variety of ceratopogonids, including *Forcipomyia*. More permanent aquatic habitats such as sugarcane leaf-axils and epiphytic bromeliads also produce potential pollinating insects (17, 19), but investigations of these types of larval habitats have been limited.

Living plants that impound water within leaf axils and flower bracts (Phytotelmata) are common in the Neotropics. They are known to provide larval habitats for many ceratopogonid species and would become increasingly important when other sites become dry. Because of their potential significance as sources of cacao pollinators and their usual abundance in cacao plantations, we conducted a special survey of phytotelmata in the experimental plots at the Cacao Research Center (CEPEC - CEPLAC) Bahia, Brazil.

## MATERIAL AND METHODS

Three types of Phytotelmata were found to be common at CEPEC:

1. *Vriesea procera* (Bromeliaceae) — one of several epiphytic tank species occurring naturally upon large shade trees (*Erythrina glauca*) in mature plots.

2. *Musa* (AAB Group). Banana prata (Musaceae) — a banana frequently cultivated with cacao and often used as shade trees in young plots.
3. *Calathea* sp. (Marantaceae) — a wild flower common in nearby, abandoned cacao plots and secondary successional forests.

All three of these plants impound various amounts of water. The bromeliad and banana have modified leaf-axils that collect water from rainfall and condensation, and the inflorescence of *Calathea* sp. secretes a mucous liquid between the bracts (Fig. 1 - 3). Collections were made between 24 March and 7 April, 1977, near the end of a 4 week period of reduced precipitation (Fig. 4) when cacao pods and other similar habitats were completely dry and generally void of immature ceratopogonids.

Ten specimens of each plant were carefully removed from the field and immediately transported to the laboratory for inspection. Bromeliads were initially emptied by immersion into a large bucket of tap water and then the leaf axils were individually washed out with a flexible hose from the tap. Banana stalks were completely dismantled and the leaf-axils were similarly flushed with water. *Calathea* sp. inflorescences were immersed in a pan of water and agitated vigorously to remove the living organisms. The rinse water from each plant

was strained through a fine mesh screen to recover the invertebrate fauna which was then separated into species groups and either preserved in 70% ethanol or reared to maturity for identification.

## RESULTS

The invertebrate communities contained in the three Phytotelmata were surprisingly complex representing 31 species and totalling 965 organisms. The structure of each community (number of individuals per species) is represented in Fig. 5 and is calculated from the total of the 10 samples. The ranks of ceratopogonid members of the communities are represented by a star.

In *Calathea* sp. and banana the invertebrate communities were either dominated or co-dominated by a ceratopogonid, but in each case by a different and new, undescribed species. *Culicoides* sp. comprised 87% of the total fauna inhabiting *Calathea* sp., and *Forcipomyia (Warmkeia)* sp. comprised 50% of the community from banana. The community inhabiting epiphytic tank bromeliads was much more diverse with *F. (Phytohelea) caribbeana* being third in abundance, but representing only 5% of the total fauna. The complete species list of each community studied appears in Table 1, along with the frequencies of occurrence.

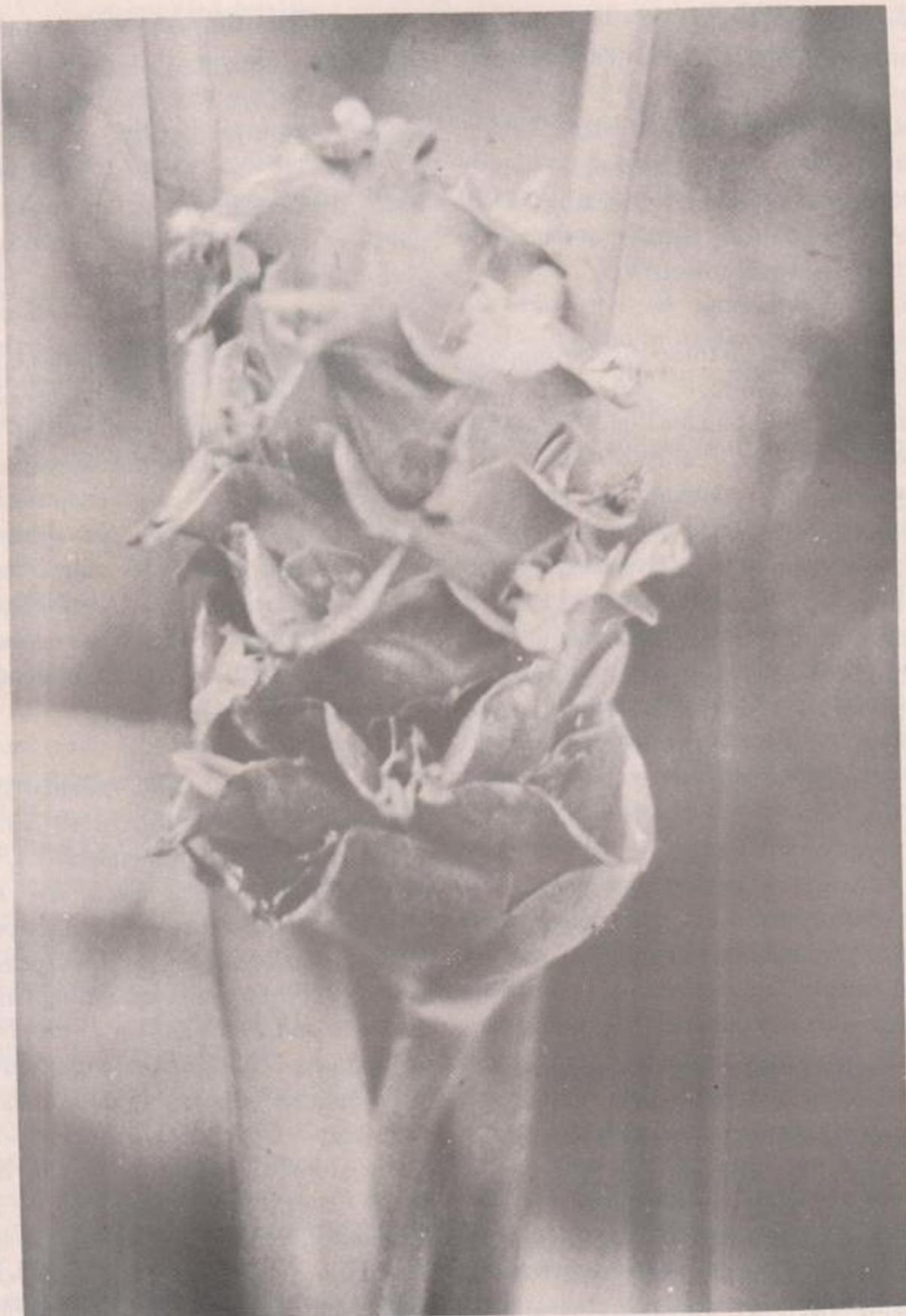


Figure 1 - Inflorescence of *Calathea* sp. which secretes a mucous liquid between the bracts.



Figure 2 – A banana stalk showing the modified leaf axils that collect rain water.

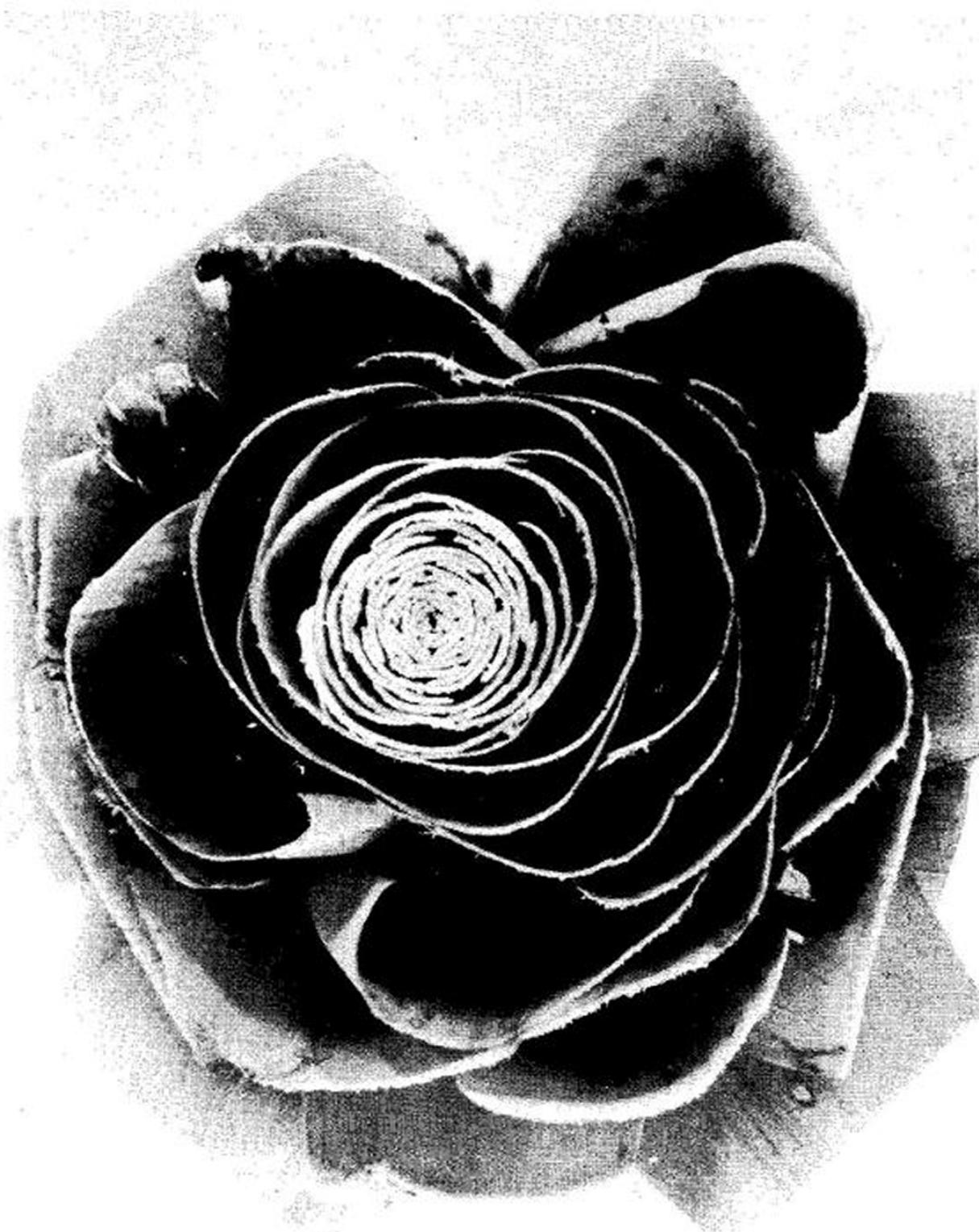


Figure 3 - Cross-section of an epiphytic tank bromeliad showing inflated leaf axils that collect rain water.

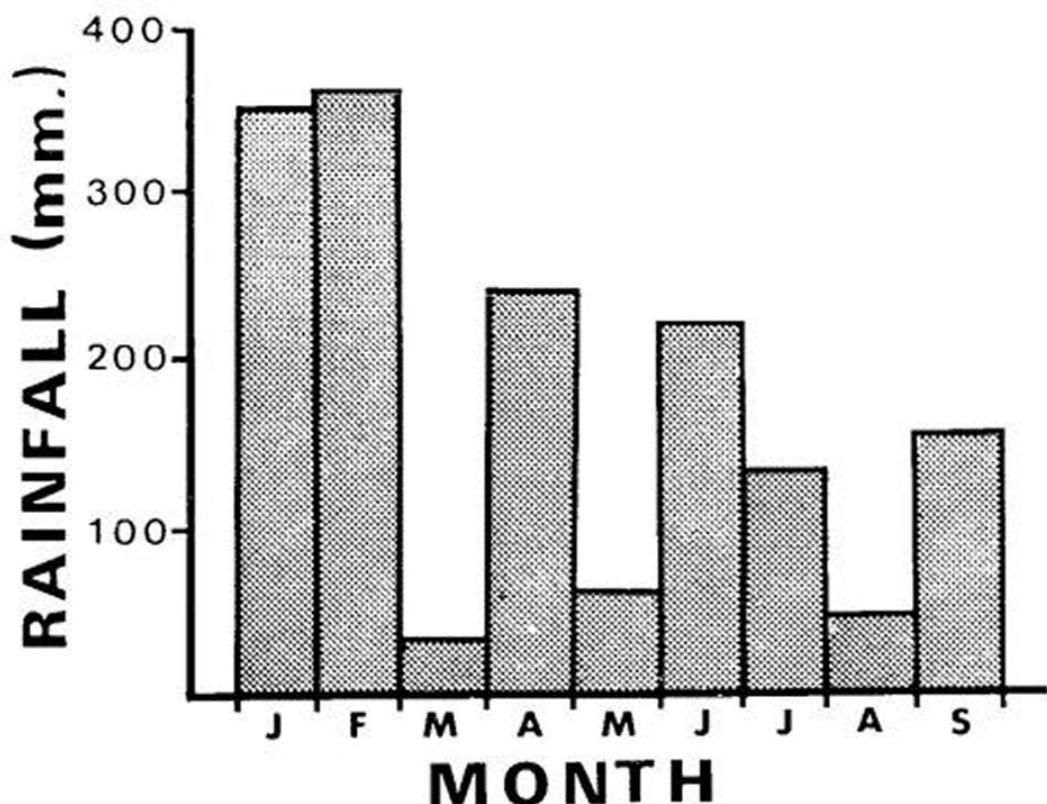


Figure 4 - Histogram of rainfall occurring during the first three quarters of 1977, Cacao Research Center, Ilhéus, Bahia, Brasil.

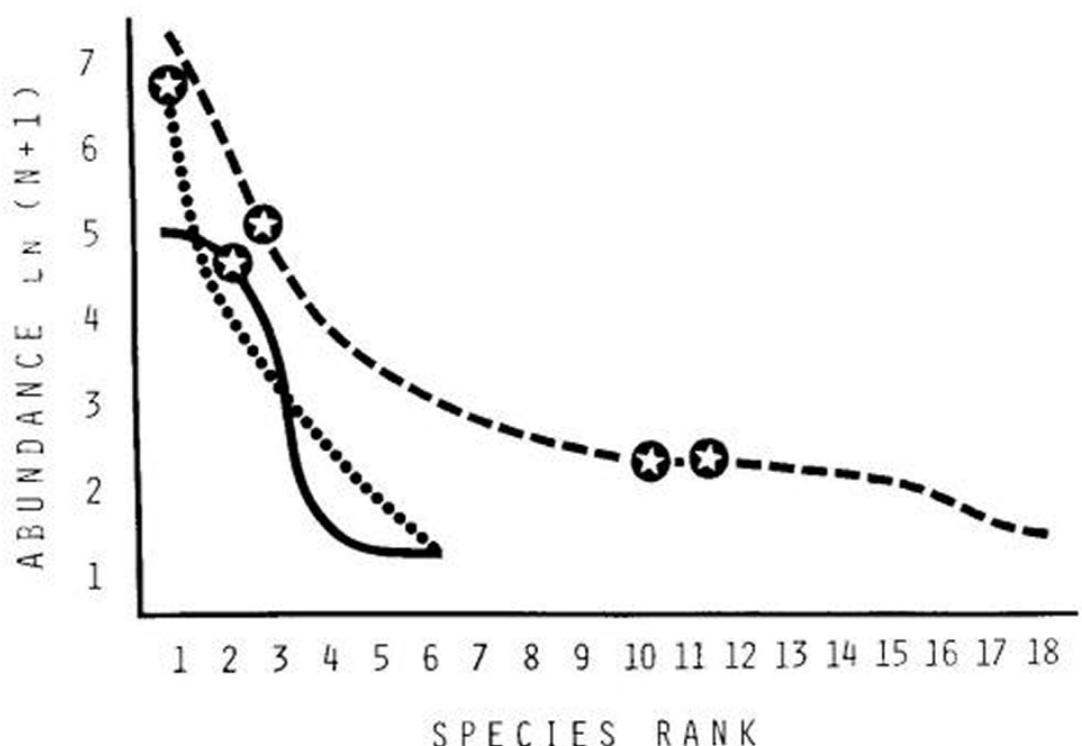


Figure 5 - The structures of the aquatic communities inhabiting bromeliads (dashed line), *Calathea* sp. (dotted line), and banana (solid line). Stars indicate the rank of ceratopogonid members in relation to the other fauna.

Table 1 - List of phytotelm inhabitants at CEPEC (sample size = 10). Ilhéus, Brazil.

<u>TAXON</u>	<u>ABUNDANCE</u>	<u>TAXON</u>	<u>ABUNDANCE</u>
<u>Vriesea procera</u> (Bromeliaceae)		<u>Calathea</u> sp. (Marantaceae)	
Oligochaeta:		Ceratopogonidae:	
Naididae	449	<u>Culicoides</u> sp.	223
Chironomidae:		Chironomidae:	
Pentaneurini	93	<u>Tanytarsini</u>	14
Ceratopogonidae:		Histeridae	11
<u>Forcipomyia</u> ( <u>Phytohelea</u> )		Psychodidae	4
<u>caribbeana</u>	30	<u>Muscidae</u> (predatory)	3
Chironomidae:		<u>Brachycera</u> (predatory)	1
<u>Tanytarsini</u>	14		
Odonata:			
Zygoptera	14		
Tipulidae	8	<u>Musa</u> (AAB Group) 'Banana prata' (Musaceae)	
Dytiscidae (larval)	7		
Culicidae:		Aulacigastriidae	33
<u>Culex</u> ( <u>Microculex</u> ) sp.	5		
Dytiscidae (adult)	4	Ceratopogonidae:	
Ceratopogonidae sp. A	3	<u>Forcipomyia</u> ( <u>Warmkeia</u> ) sp.	24
sp. B	3		
Culicidae:		Brachycera sp. A	5
<u>Wyeomyia</u> sp.	3	Annelida:	
<u>Corethrella</u> sp.	3	<u>Hirudinea</u>	1
Oligochaeta	2	Oligochaeta	1
Brachycera	2	Brachycera sp. B	1
Anura	2	Culicidae:	
Hydrophilidae	1	<u>Sabethini</u>	1
Aulacigastriidae	1		

## DISCUSSION

All three Phytotelmata proved to be favorable habitats for immature ceratopogonids. It was surprising that the relatively small inflorescence of *Calathea* sp. would produce such a large number of *Culicoides* sp. (22.4 ave.). This plant may well be the only larval habitat for this insect as it has never been collected before despite several years of intensive collecting of ceratopogonids in CEPEC. Other species of *Calathea* might support different ceratopogonid species as Wirth (personal communication) found *Forcipomyia* sp. in *C. lutea* in Dominica. In addition, the water-holding flower bracts of other plants deserve investigation, such as *Heliconia* (Musaceae) and several species of Zingiberaceae which are known breeding sites for ceratopogonids (18, 24).

In banana leaf-axils, *Forcipomyia* (*Warmkeia*) sp. was second in abundance, but averaged only 2.4 larvae per plant. The dominant member of this community was a predatory aulacigastrid fly, probably *Stenomicra* sp.. It is difficult to explain the dominance of a predator in a community, but most specimens were near maturity indicating that the incidence of its presumed prey, *F. (Warmkeia)* sp., might have been greater at an earlier time. Even at low densities, the total numbers of ceratopogonids produced by banana leaf-axils would be considerable when many plants are culti-

vated in or near cacao and especially when they are used as shade trees.

Rotting banana stems have been previously investigated as ceratopogonid breeding sites and at least four species have been found (17, 19), but not *F. (Warmkeia)* sp.. Apparently this species is restricted to living plants. According to Saunders (11), the entire subgenus *Warmkeia* is commonly found in plant axils.

The epiphytic tank bromeliad contained a slightly higher density of ceratopogonid larvae of 3.6 per plant even though *F. (Phytohelea) caribbeana* was the third most abundant invertebrate in this community. An oligochaete worm and a chironomid were more abundant. Three specimens each of two additional ceratopogonid species were also found (Fig. 5, Table 1), but failed to reach maturity and could not be identified.

The densities attained by epiphytic tank bromeliads upon shade trees in cacao plantations make them extremely important as larval habitats for ceratopogonids as well as other microdiptera. Pittendrigh (10) reports densities as high as 155 bromeliads per shade tree (*Erythrina micropteryx*) in cacao plantations in Trinidad, composed almost exclusively of water-holding species. No estimates were made of the bromeliad densities at CEPEC, but 10 bromeliad specimens was an exceedingly small sample size in relation to the total bromeliad flora and only one species was sampled.

In previous studies at CEPEC, at least 16 species of ceratopogonids, including seven subgenera of *Forcipomyia*, have been collected from a total of 11 epiphytic bromeliads (19, 20). These plants are known to be larval habitats for many ceratopogonid species throughout the Neotropics (2, 9, 12, 21, 22, 23, 24) and in the U.S. Virgin Islands they comprise 32% of the total bromeliad fauna (8). In view of their abundance in cacao plantations in the Neotropics, bromeliads certainly deserve more consideration as potential larval habitats for pollinating insects.

With the possible exception of *Calathea* sp. which supplies its own fluid medium for the development of aquatic fauna, most Phytotelmata are subject to fluctuating environmental conditions such as rainfall and humidity, and the inhabiting aquatic community will experience variation in both structure and species composition (2). Banana plants and especially bromeliads should be surveyed throughout the year and in larger numbers in order to properly assess their significance as larval habitats of ceratopogonids.

The results of this study indicate that there is a certain degree of host-plant specificity among ceratopogonids since three different species were collected from three unrelated plant habitats in the same area with no overlap in occurrence. If each type of phytotelm habitat is occupied by a

distinct ceratopogonid species, then further investigations of other similar habitats should reveal a more diverse ceratopogonid fauna than is generally recognized to exist in association with cultivated cacao.

It is evident from the variety of insect species that have been reported to be pollinators of cacao, that existing pollination success is related to the abundance and diversity of whatever suitable insects happen to be in the immediate area. Increasing the abundance and diversity of larval habitats such as Phytotelmata, which are here indicated to be very favorable for ceratopogonids, may substantially change the adult insect populations and increase the chances of more species participating in the pollination process. Although the kinds and numbers of insects produced by these plants will vary seasonally, it is apparent that they are continuous sources of a variety of ceratopogonids, even in dry weather when pollination rates are low.

However, caution is advised in any attempts to manipulate Phytotelmata populations, especially bromeliads. Although they may be good sources of pollinating insects, they are also excellent sources of insects of medical importance such as bromeliad-inhabiting mosquitoes, tabanids, and blood-feeding *Culicoides*. *Anopheles (Kerteszia)* mosquitoes have caused severe malaria outbreaks among cacao workers in both Trinidad and Brazil in the past (1, 13).

Maximizing the production of insect pollinators inhabiting bromeliads and other phytotelmata without also increasing the risk of human disease can only be achieved with more detailed study of the insect-plant relationships among these unique plants.

## CONCLUSIONS

1. Water-holding plants (Phytotelmata) provide favorable larval habitats for ceratopogonids during dry weather in Bahia, Brazil.

2. Ceratopogonids rank high in the structures of communities contained in Phytotelmata and are sometimes dominant.

3. Each type of phytotelm habitat may support specific ceratopogonid species, resulting in overall increased species diversity of potential pollinating insects in the vicinity of cacao plantations.

4. More studies are needed on insect-plant relationships of Phytotelmata in order to fully assess their potential usefulness in increasing cacao pollination.

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## RESUMO

### Plantas Retentoras de Água (Phytotelmata) como Habitats de Larvas de Ceratopogonídeos Polinizadores do Cacaueiro na Bahia, Brasil.

Ao final de um período seco no Sul da Bahia, Brasil, 10 amostras de três espécies de plantas retentoras de água (Phytotelmata) foram examinadas à procura de larvas de ceratopogonídeos (Diptera), polinizadores potenciais do ca-

cacau. A flor silvestre do *Calathea* sp. (Marantaceae) abrigava uma média de 22,4 larvas de *Culicoides* sp. na água inter-axilar da sua inflorescência. A bananeira *Musa* (Grupo AAB) banana prata (Musaceae) abrigava, na água retida nas axilas foliares, uma média de 2,4 larvas de *Forcipomyia (Warmkeia)* sp. Um gravatá tanque epífita *Vriesea procera* (Bromeliaceae) abrigava uma média de 3,0 larvas de *F. (Phytohelea) caribbeana* também nas axilas foliares cheias de água. Os ceratopogonídeos em geral foram dominantes na comunidade *Calathea*, co-dominantes em banana e menos abundantes nos gravatás. Phytotelmata associados com cacau cultivado são indicados como habitats favoráveis para determinadas espécies de ceratopogonídeos. Incrementando a diversidade das plantas armazenadoras de água, é possível, consequentemente, incrementar as oportunidades de mais espécies de insetos participarem no processo da polinização do cacaueiro, particularmente durante o período seco. Todavia, o potencial dos gravatás como fonte de insetos de importância médica, tais como os mosquitos vetores de malária, deve ser também considerado, quando se pretender manipular a flora Phytotelmata nas plantações de cacau, com o intento de aumentar as taxas de polinização.



**ANTAGONISMO AO FUNGO  
*Crinipellis perniciosa* (Stahel) Singer,  
CAUSADOR DA VASSOURA-DE-BRUXA DO CACAUEIRO**

*Cleber Novais Bastos\**

**ABSTRACT**

**Antagonism to the Fungus *Crinipellis perniciosa* (Stahel) Singer  
Causal Agent of Witches' Broom Disease of Cacao**

In the present study the antagonistic capacity of six isolates of fungi and one bacterial isolate against *Crinipellis perniciosa* (Stahel) Singer was evaluated.

The tests were conducted *in vitro* and the antagonistic capacity was determined by the measuring of inhibition zones. All the isolates tested had an inhibitory effect on growth of *C. perniciosa* with *Eurotium* sp. producing the greatest inhibition (22.2 mm) and *Aspergillus giganteus* least (4.0 mm).

**INTRODUÇÃO**

O fenômeno de antagonismo entre microrganismos ocorre tanto em laboratório como na natureza e, por isso, tem merecido a atenção dos pesquisadores, sobretudo dos fitossanitaris-

tas, visando à sua utilização no controle biológico das doenças de plantas.

Na literatura, são encontradas inúmeras citações de trabalhos sobre antagonismo de microrganismos a fungos fitopatogênicos (1, 2, 4,

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5, 6, 7, 9), inclusive basidiomicetos (3, 8). Entretanto, não existe referência que assinale a constatação de antagônicos a *Crinipellis perniciosa* (Stahel) Singer (*Marasmius perniciosus* Stahel).

O presente trabalho teve por objetivo detectar e selecionar microrganismos antagônicos ao *C. perniciosa*, na tentativa de utilizá-los no controle biológico da vassoura-de-bruxa do cacau.

## MATERIAIS E MÉTODOS

Foram utilizados sete isolados, sendo seis de fungos e um de bactéria. Os de fungos foram: *Penicillium lilacinum* Thom, *P. purpureescens* Sopp., *Eurotium* sp., *Aspergillus giganteus* Whemer, *Penicillium* sp. (contaminante de culturas de *C. perniciosa*) e *A. terreus* (Bloch) Thom & Raper (parasita de basidiocarpos de *C. perniciosa*). O de bactéria foi *Bacillus subtilis* Cohn (contaminante de culturas de *C. perniciosa*). Os quatro primeiros são fungos antagônicos a *Phytophthora palmivora* (Butl.) Butl., conforme demonstrado por Figueiredo\*.

Os testes para se determinar a capacidade antagônica foram conduzidos em placas de Petri contendo extrato

de malte-agar, sendo o antagônico e o *C. perniciosa* colocados, simultaneamente, em bordos opostos das placas, distanciados de 6 cm. Para cada tratamento foram feitas quatro repetições.

As placas foram mantidas à temperatura ambiente e, após 12 dias, foi determinada a zona de inibição (distância em mm da borda da colônia antagônica à da colônia do *C. perniciosa*).

## RESULTADOS E DISCUSSÃO

Resultados preliminares da inibição de *C. perniciosa* pelos microrganismos são apresentados no Quadro 1. Observa-se que todos os isolados testados demonstraram atividades antagônicas, destacando-se, porém, *Eurotium* sp., por produzir maior zona de inibição (22,2 mm). Por outro lado, *A. giganteus* foi o que demonstrou menor grau de antagonismo (4,0 mm).

Quadro 1 - Efeito inibidor de alguns microrganismos antagônicos a *Crinipellis perniciosa* (Stahel) Singer *in vitro*.

Antagônicos	Zona de Inibição (mm)
<i>Penicillium lilacinum</i>	15,2
<i>Penicillium purpureescens</i>	15,0
<i>Aspergillus terreus</i>	12,2
<i>Eurotium</i> sp.	22,2
<i>Penicillium</i> sp.	11,7
<i>Bacillus subtilis</i>	12,7
<i>Aspergillus giganteus</i>	4,0

\* FIGUEIREDO, J.M. Dados não publicados. Divisão de Fitopatologia, Centro de Pesquisas do Cacau, C.P. 7, 45.600, Itabuna, Bahia, Brasil.

*A. terreus*, isolado como parasita de basidiocarpos de *C. perniciosa* comprovou sua ação antagônica mesmo depois de repicado para meio de cultivo.

A Figura 1 mostra que o antagônico de maior atividade, *Eurotium* sp., apresentou menor desenvolvimento da colônia, sugerindo que a inibição produzida em *C. perniciosa* seja mais consequência de antibiose do que

exaustão de fator de crescimento. Assim, acredita-se que a zona de inibição produzida *in vitro* seja decorrente da produção de substâncias pelos antagonistas, que impedem o desenvolvimento do patógeno (9).

Os resultados desse trabalho mostram apenas um teste preliminar de antagonismo. Novos ensaios deverão ser realizados para confirmá-los e elucidar a natureza do efeito antagônico ao *C. perniciosa*.

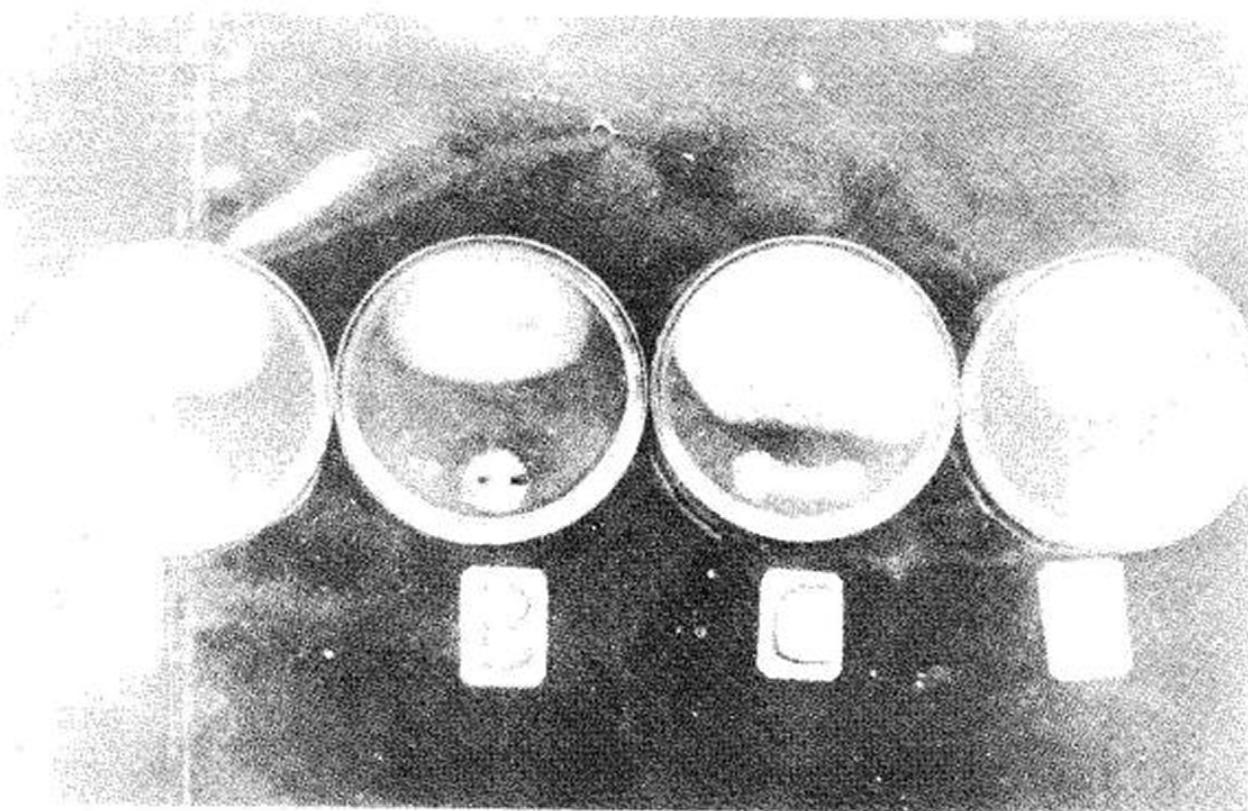


Figura 1 - Inibição de *Crinipellis perniciosa* pelos antagônicos: A - *Penicillium lilacinum*; B - *Eurotium* sp.; C - *Bacillus subtilis*; D - *Penicillium* sp.

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## RESUMO

No presente trabalho foi determinada a ação de sete isolados de fungos e um de bactéria sobre *Crinipellis perniciosa* (Stahel) Singer.

Os testes foram realizados *in vitro* e a atividade antagônica foi determinada através da mensuração da zona de inibição. Todos os isolados testados produziram efeito inibitório sobre o *C. perniciosa*, destacando-se *Eurotium* sp. Por produzir maior zona de inibição (22,2 mm) em contraste com o de menor ação, *Aspergillus giganteus* (4,0 mm).



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