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Order Number 9304726

Endophytic fungi in the tropical palm *Euterpe oleracea* Mart

Rodrigues, Katia Ferreira, Ph.D.

City University of New York, 1992

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ENDOPHYTIC FUNGI IN THE TROPICAL PALM *EUTERPE OLERACEA* MART.

by

KATIA FERREIRA RODRIGUES

A dissertation submitted to the Graduate
Faculty in Biology in partial fulfillment
of the requirements for the degree of Doctor
of Philosophy, The City University of New York

1992

This manuscript has been read and accepted for the Graduate Faculty in Biology in satisfaction of the dissertation requirement for the degree of Doctor of Philosophy.

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ABSTRACT

ENDOPHYTIC FUNGI IN THE TROPICAL PALM *EUTERPE OLERACEA* MART.

by

Katia Ferreira Rodrigues

Chairman: Gary J. Samuels

The occurrence of foliar endophytes in the Amazonian palm *Euterpe oleracea* Mart. was investigated during January and September of 1989 and 1990. Endophytic fungi were isolated from leaflets of ten 6-to 18-month-old seedlings, and of ten 5-to 20-year-old trees located at the Combu Island, a river-island located in the Brazilian Amazon estuary near Belém. Endophytic colonization frequencies were positively correlated with leaf age, plant developmental stages, and subsite. Mature leaves showed higher fungal colonization than the young ones; seedlings were more intensely colonized than trees. Community ordination analysis (MDS) has shown that trees and seedlings located at different subsites were colonized by distinct endophytic communities.

Fifty-seven endophytic species and six familial taxa were identified, *Xylaria cubensis* and *Letendraea* sp. were the most dominant colonizers. The endophytic fungi isolated could be generally distinguished by two categories, one representing those taxa that have been primarily referred to as saprophytes, and another category comprising species that have been previously recorded as pathogens in taxonomically close related host plants. Some of the fungi isolated herein represent new species, viz. *Idriella amazonica*, *Idriella asaicola*, *Idriella euterpes*, and *Letendraea* sp.

Eighty-one isolates representing thirteen species of *Xylaria* were examined for isozyme variation using starch gel electrophoresis of initially twenty-one enzymes. Five enzymes were selected which represented the most clear resolution. Results from the isozyme analysis revealed a high degree of intra- and interspecific diversity among the thirteen *Xylaria* species.

A combination of biochemical and cultural data were used to characterize thirty-five endophytic isolates of *Xylaria cubensis* complex, and then subjected to numerical analysis. In addition, vegetative compatibility group analysis were carried out with the same 35 isolates of *X. cubensis* in order to assess genetically different (vegetatively incompatible) individuals colonizing the same and different leaves. All pairings of an isolate with itself were compatible. However, several strains were vegetatively incompatible with all other strains, showing that different populations of *Xylaria cubensis* are present in a single tree.

ACKNOWLEDGMENTS

I would like to express my profound appreciation to Dr. Gary J. Samuels for his patience, friendship, and unparalleled guidance. His meticulous suggestions throughout each step of my research, and his valuable contribution toward the fulfillment of this manuscript, including his help with the identification of species that were hard to identify were appreciated. I am also deeply indebted to Dr. Orlando Petrini (Mikrobiologisches Institut-ETH, Zürich, Switzerland) for his many enlightening discussions on endophyte research, and helpful assistance concerning the methodology, vegetative compatibility analysis and statistical evaluation of the data. The opportunity that he gave me to spend two months in his laboratory deepened the significance of the research through the addition of physiological, macromolecular, and genetic aspects of the host-endophyte symbiosis. His valuable review of the manuscript is greatly appreciated. I also wish to thank the members of my Graduate Committee, Drs. Amy Rossman, Andrew Henderson, Dennis Stevenson, and Dwight Kincaid for their willingness to share their knowledge.

The suggestions and guidance of several other mycologists were also greatly appreciated. Dr. Liliane Petrini offered helpful comments about taxonomy of some xylariaceous fungi, and warm hospitality during my visit in Switzerland. Dr. Clark T. Rogerson, as always, stood ready with logistical and moral support throughout the course of this research. Prof. Jack Rogers and Dr. Brenda Callan offered advise about the taxonomy of xylariaceous fungi and contributed with living cultural collections. I am also indebted to

Prof. Jack Rogers for allowing me laboratory space and sharing his time for two weeks at Washington State University. Dr. Adrian Leuchtmann (Geobotanisches Institut-ETH, Zürich, Switzerland) guided through the isozyme analysis in his laboratory; he was very helpful and generous with his expertise. Dr. Ignacio Chapela (Sandoz Ltd, Basle, Switzerland) kindly reviewed earlier versions of this manuscript.

I am deeply indebted to the following collaborators who gave their time and advise: Dr. Anthony Anderson (Museu Paraense Emilio Goeldi, Belém, Brazil) facilitated the physical and technical support in the collection of leaf samples at Combu Island. Dr. Rupert Barneby shared his expertise on botanical Latin. Dr. Fernando Carneiro de Albuquerque and Dr. Emeleocípio Botelho de Andrade, the director of EMBRAPA/CPATU without which this work would have been very difficult. Mrs. Gabi Leuchtmann was very helpful in assuring a pleasant stay.in Zürich.

I am extremely thankful to the New York Botanical Garden and Texaco Philanthropic Foundation for awarding me a Herbarium Graduate Fellowship. National Science Foundation Doctoral Dissertation Improvement Grant BSR-8914564 to G. J. Samuels and myself partially supported this investigation.

I thank the following institutions which provided technical assistance and partial financial support during my laboratory work: USDA, Systematic Botany and Mycology Laboratory, especially to Dr. Amy Rossman, Miss Upasana Bhatnagar and Anita Phillips; Swiss Federal Institute of Technology, ETH-Zentrum, especially to Drs. O. Petrini, A. Leuchtmann and Thomas Sieber, along with Mr. L. Toti and O. Viret.

Finally, I would like to thank my husband Moacyr, for his support and encouragement. My father, my sister, and my mother- and father-in-law always encouraged my studies.

I would like to dedicate this work to the memory of my mother.

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APPENDICES

1. INTRODUCTION

1.1. Biology of endophytes

Living plants have been shown to harbor fungi inside their tissue, and the presence of the fungi may affect the host in different ways. Associations between plants and fungi in which the participants seems to gain some benefits are considered mutualistic. These associations include mycorrhizae and endophytes. Mycorrhizal associations refer to the fungi integrated in the plant root system, playing an important role in the life cycle of a plant by enhancing root uptake of nutrients from the soil (Harley 1989). The term "endophyte" as originally used by De Bary (1866) refers to all organisms occurring within plant tissue. Carroll's (1986) definition denotes organisms that colonize aerial parts of living plant tissues, not inducing visible signs of disease, i.e., causing asymptomatic infections within healthy stems and leaves. Pathogenic fungi and mycorrhizae are excluded from his concept. Petrini (1991) proposes an expansion of Carroll's definition, including all organisms living within plant tissues, and incorporated into his concept are the latent pathogens which are now known to live symptomlessly inside their hosts during part of their life cycle. In this dissertation the term "endophyte" is used in a broad sense to include those fungi that spend all or nearly all of their life cycle in the host plant tissue (sensu De Bary).

1.2. Colonization and detection of endophytes

Three possible modes of leaf colonization by fungal endophytes have been proposed by Petrini (1991), viz. air spora, seeds, and insects. The first has been considered the most widespread mechanism of colonization of aerial plant tissues employed by endophytes. The second has been shown to be the only means of dissemination of *Acremonium* in grass endophytes (Siegel et al. 1987a) and is a way of assuring their transmission through succeeding host generations. The third, although not yet conclusively demonstrated, was suggested based upon the strong fungus-insect interactions known to occur with tree pathogens (Sinclair et al. 1987).

The isolation and identification of endophytic fungi is usually accomplished by a rigorous surface sterilization of the plant tissue followed by incubation on culture media (Petrini 1986). Despite studies based on cultural work alone, other techniques have been used in order to determine endophytic fungal infections inside the host tissue. These include microscopic as well as biochemical techniques. Studies on endophytic fungi based on electron microscopy observations (Bernstein and Carroll 1977, Hinton and Bacon 1985; Siegel et al. 1987b; Stone 1988; Suske and Acker 1987; 1990), immunoelectron microscopy techniques (Suske and Acker 1989; 1990), and enzyme-linked immuno-sorbent assays (ELISA) [Johnson et al. 1982] have been reported.

The vegetative hyphae produced by fungal endophytes have been reported to grow either intercellularly, or intracellularly within the host plant tissues. In grass endophytes the hyphae grow strictly in a intercellular fashion, running longitudinally through leaves and stems as reported by Siegel et al. (1987b) and Clay (1989) from *Festuca* and *Lolium* species. Results from histological investigations have shown that endophytic colonization in conifer needles is characterized by the production of intracellular hyphae that are confined to single epidermal cells as reported by Stone (1987; 1988) in coastal redwood and Douglas fir [*Sequoia sempervirens* (D. Don ex Lamb.) Endl. and *Pseudotsuga menziesii* (Mirb.) Franco , respectively]. In general, very few endophytic species have been seen inside their host tissues, much less localized.

Little is known about fungal endophyte biomass. According to Siegel et al. (1984) endophyte biomass in tall fescue grass (*Festuca arundinacea* Schreber) was less than 1% of the total plant biomass. Carroll (1991) estimated the endophyte biomass of *Rhabdocline parkeri* needles to be around 0.01-0.1%. Based on this information Carroll (1991) proposed that, when compared to conifers, a higher metabolic cost to the plant might be expected in grass endophytes associations.

1.3. Endophytes of grasses

1.3.1. Clavicipitaceous endophytes

A great number of investigations on fungal endophytes of grasses (Poaceae) and sedges (Cyperaceae and Juncaceae) have been reported (see review in Clay 1986; 1988 and Siegel and Schardl 1991). The effects of the endophytes on plant fitness (Latch et al. 1985; Clay et al. 1989) and alkaloid production support the assumption of a mutualistic association with their hosts. This association has been referred to as a "constitutive mutualism", in which the endophyte develops a systemic infection within the plant tissues that is carried to a new plant through the seeds (Carroll 1988). These systemic infections occur in the leaves, culms and inflorescences. They are caused by members of the Clavicipitales (Ascomycotina), family Clavicipitaceae, tribe Balansieae, and include species of the genera *Atkinsonella*, *Balansia*, *Epichloë*, and *Myriogenospora.* The asexual states of the Balansieae comprise species belonging to the form-genus *Acremonium* section *Albo-lanosa*, and most of these fungi are referred to as "true endophytes" (Siegel et al. 1987; Siegel and Schardl 1991), i.e. they do not produce disease in the plant associate. This mutualistic association involves endophytes that do not produce stromata on infected plants, being found in festucoid grasses including *Festuca arundinacea* Schreb., *F. versuta* Beal., *Lolium perenne* L., *L. temulentum* L., *Stipa eminens* Cav., and *S. robusta* (Vasey) Scribn. Conversely, species of *Epichloë*, *Atkinsonella* and *Balansia* have been reported to produce stromatic tissue around the inflorescences of host individuals belonging to several subfamilies of the Poaceae and in some sedges (White 1988). This type of association is referred as "choke disease" of grasses and is classified as pathogenic because of the negative effect on host plants.

The benefits provided by clavicipitaceous endophytes to their hosts have generally been divided into two broad categories. First, the endophyte modifies the host plant physiology by enhancing plant growth (Latch et al. 1985; Clay 1987; West et al. 1988) and increasing resistance to abiotic stress (Belesky et al. 1987; Arachevaleta et al. 1989). Second, the production of toxic alkaloids by the endophyte is known to increase the plant resistance to herbivory by grazing mammals (Bacon et al. 1986; White 1987; Bacon and Siegel 1988; Clay 1988; Cheplick and Clay 1988; Siegel and Schardl 1991), insects (Prestidge et al. 1982; Clay et al. 1989; Siegel et al. 1990), and nematodes (Pedersen et al. 1988; West et al. 1988).

As a result of their seed-borne nature, these endophytes have been carried along with their hosts and have become worldwide in distribution.

1.3.2. Non-clavicipitaceous grass-endophytes

Investigations on asymptomatic infections in grasses caused by endophytes unrelated to the clavicipitaceous fungi have been reported. Latch et al. (1984) found endophytic taxa referred to as *Gliocladium*-like and *Phialophora*-like, respectively in ryegrass (*Lolium perenne* L.) and tall fescue (*Festuca arundinacea* Schreber). White et al. (1990) documented the presence of a seedborne non-clavicipitaceous endophyte, *Pseudocercosporella trichachnicola* White & Morgan-Jones, in a warm-season perennial grass [*Trichachne insularis* (L.) Nees]. Sieber et al. (1988) reported over 200 endophytic species from winter wheat (*Triticum aestivum* L.) cultivars, and Fisher and Petrini (1992) reported over 30 species endophytic in rice (*Oryza sativa* L.) cultivars. Sieber et al. (1988) and Fisher and Petrini (1992) demonstrated the presence of plant pathogens among the endophytes in apparently healthy tissues. The possible antiherbivore effects of non-clavicipitaceous endophytes in grasses are still under investigation (Clay 1990).

1.4. Endophytes of woody angiosperms

A large number of investigations on symptomless infections in angiosperms other than grasses, caused by endophytic fungi have been reported from various host families and localities (summarized in Petrini 1986 and in Boddy and Griffith 1989). However, endophytes of the Ericaceae are possibly the most thoroughly studied (Petrini et al. 1982; Petrini 1984; Widler and Müller 1984; Petrini 1985; Petrini 1987). Surveys of endophytic fungal communities of several other host families including the Asteraceae (Cabral et al. 1990), Betulaceae (Fisher and Petrini 1990), Chenopodiaceae (Fisher and Petrini 1987a), Fagaceae (Chapela 1989; Sieber and Hugentobler 1987), Fabaceae (Fisher and Petrini 1987b), and Myrtaceae (Bertoni and Cabral 1988) have also been reported. Most of these studies have been carried out in temperate (see Petrini 1986; 1991) and subtropical regions (Argentina: Cabral 1985; Bertoni and Cabral 1988; Faifer and Bertoni 1988). There are only few reports from tropical regions (Petrini and Dreyfuss 1981; Dreyfuss and Petrini 1984; Rodrigues and Samuels 1990; 1992).

Investigations of species composition of endophytic communities have shown variations in infection frequencies to be influenced by factors such as leaf age and seasonal change. It was demonstrated that old leaves tended to support more species and/or greater frequency of colonization than young leaves. This has been reported for *Ulex europaeus* L. and *U. gallii* Planch. (Fisher et al. 1986; Sieber and Hugentobler 1987), and *Eucalyptus viminalis* Labill. (Bertoni and Cabral 1988). Studies emphasizing seasonal variation have yielded conflicting results. In *Arctostaphylos uva-ursi* (L.) Sprengel the most common endophytes were recovered throughout the year as reported by Widler and Müller (1984), whereas in beech leaves (*Fagus* *sylvatica* L.) Sieber and Hugentobler (1987) detected seasonal variations in colonization rates by the most frequent endophytes.

The presence of endophytes within woody stems have been shown to be as common as endophytes within leaves (Chapela 1989; Chapela and Boddy 1988a; 1988b; Fisher et al. 1986; Petrini and Fisher 1988; Sieber 1989). The role of those endophytes within healthy wood has been associated with the protection of stems against attack by insects and fungi, as well as with the abscission process of dead branches (Carroll 1991). Chapela and Boddy (1988a; 1988b) have shown that the most common wood endophytes of beech (*Hypoxylon* spp., *Nectria coccinea*, *Coniophora puteana*) and aspen (*Hypoxylon* spp., *Cryptosphaeria populina*, *Cytospora chrysosperma*) live in a latent form for several years, and the onset of infection is positively correlated to the water contents of the sapwood, i.e., under drought conditions the fungi are released and colonize the wood.

Contrary to the clavicipitaceous endophytes, endophytic fungi of woody plants are rarely transmitted through seeds, forming a much looser association with their hosts. This type of mutualistic association is known as "inducible mutualism" (Carroll 1988).

1.5. Endophytes of gymnosperms

Fungal endophytes of conifers have been extensively studied by Carroll et al. (1977), Carroll and Carroll (1978), Petrini and Carroll (1981), Stone (1986; 1987; 1988), Sieber-Canavesi and Sieber (1988), Sieber (1988), Suske and Acker (1989), Espinosa-Garcia and Langenheim (1990), Sieber-Canavesi et al. (1991).

Several studies have shown that factors such as location and humidity of the site and leaf age are highly correlated with colonization rates presented by endophytic fungi. It has been demonstrated that the frequency of endophytic fungi in conifers is higher in wet sites and low elevations as reported by Carroll and Carroll (1978), Petrini and Carroll (1981), Petrini et al. (1982) and Sieber (1988). It was observed that endophytic frequencies were higher in homogeneous stands with a closed canopy as reported by Petrini and Carroll (1981), Petrini et al. (1982) and Legault et al. (1989). Increase in frequency of colonization with leaf age has been reported by Petrini and Carroll (1981), Stone (1986), Sieber-Canavesi and Sieber (1988) and Espinosa-Garcia and Langenheim (1990).

Reports on endophytic fungi from gymnosperms have also provided evidence of antagonism towards insect pests. For instance, *Rhabdocline parkeri* Sherwood, a ubiquitous endophyte of Douglas fir needles, was shown to be responsible for the decline of the resident larvae of gall midges that often attack the needles (Carroll 1986). A similar case was reported by Clark et al. (1989) where endophytes of balsam fir needles (*Abies balsamea* L.) and red spruce (*Picea rubens* Sarg.) exhibited a toxic effect against spruce budworm "in vitro". The identification of the compounds active against spruce budworm larvae was recently reported by Calhoun et al. (1992).

1.6. Perspectives in endophyte research

The amount of research on endophytic fungi has greatly increased due mainly to their high biological potential. However, numerous aspects of the ecological and physiological processes governing the fungus-plant association remain unknown. For instance, the biochemical basis of the host-endophyte symbiosis, and the mechanisms by which asymptomatic infections are maintained are not known.

Because endophytic fungi are adjusted to the host plant system they have attracted both fields of biocontrol and genetic engineering of plants. The use of fungi as vectors of foreign genes into economically important crops (Wood 1990), or in biological control of pests and diseases due to their ability of producing antibiotic substances (Petrini 1991) has become a promised field for applied research explorers. In addition, antibiotic compounds produced "in vitro" by endophytic fungi have been reported to have an active effect against human and plant pathogenic bacteria (Fisher et al. 1984a; 1984b; Dreyfuss 1986).

1.7. Proposed research

The primary goal of this research was to demonstrate the presence of endophytic fungi within symptomless leaves of *Euterpe oleracea* Mart. (Palmae), and to present a comprehensive guide for identification in culture of the endophytes. The first part of this investigation will focus on the colonization frequencies of the endophytic fungi in relation to different factors such as host tissue (vein vs. intervein), plant developmental stage (tree vs. seedling), leaf age (unopened, newly expanded and mature leaves), subsite (low vs. high várzea), and season (wet vs. dry). It will also include observations on different distribution patterns presented by the most frequently isolated endophytes. The second part of the present work will deal with the taxonomy of the endophytic fungi.

1.7.1. Taxonomic considerations on *Xylaria* species and isozyme analysis

Species identification of suspected *Xylaria* species grown in culture remains a difficult problem because teleomorphs are infrequently formed. Although the identification of the anamorphs produced in culture is relatively easy due to their typical conidiophores and conidial development, their usefulness for taxonomy at generic and at the specific levels has proved to be of limited use. The majority of reports on *Xylaria* species in culture were based on specimens collected in the temperate regions. Only recently have a few tropical species been reported as growing in pure culture (Rogers et al., 1987; 1988; Rogers and Callan 1990). However as the number of specimens of individual species cultured was low, the range of variability that is expected to occur within a species is as yet not known. Under these circumstances, only cultural morphology criteria would be inadequate for the taxonomic identification, for this reason isozyme gel electrophoresis was chosen as a supplementary method.

Isozymes (or isoenzymes) are different molecular forms of a single enzyme system. These are detected by means of a separation procedure known as electrophoresis wherein different forms of individual enzyme systems migrate through a gel to which an electric charge has been applied (Murphy et al. 1990). Each molecular form migrates a specific distance in the gel over a specific time. Gels are stained for the enzyme system in question and a characteristic banding pattern results that reveals the location of the isozyme. Isozyme analysis has greatly been utilized by mycologists and plant pathologists. This technique has shown a high capability to solve taxonomic problems within several fungal taxa (Leuchtmann and Clay 1990; Bonde et al. 1991; Oudemans and Coffey 1991; Leuchtmann et al. 1992), to identify unknown fungal strains (Keller 1992), and to study population genetics (Micales et al. 1986).

Isozyme analysis was undertaken by means of starch gel electrophoresis in order to fulfill the following objectives: 1) to evaluate isolates of *Xylaria* species for isozyme activity; 2) to determine the extent of intra- and interspecific variation between species and strains; 3) to establish a suitable electrophoretic methodology for *Xylaria* species.

1.7.2. Vegetative Compatibility Group test (VCG)

Mycelial (somatic) compatibility is usually associated with vegetative compatibility (VC), and it has been widely reported in studies of population structure (Anagnostakis 1984; Bevercombe and Rayner 1984; Bosland and Williams 1987; Zambino and Harrington 1990). The occurrence of vegetative compatibility is indicated by the intermingling (anastomosis) of hyphae in culture of different strains of one taxon upon pairing, thus identifying the strains as genetically identical "clones" (Childs 1963). Conversely, the occurrence of an interaction zone where either a distinct reaction line (usually a pigmented line), or a band of abundant aerial mycelium is formed indicates genetic difference between strains.

Vegetative compatibility as defined by Puhalla (1985) is the ability of two strains to form a heterokaryon when paired. A heterokaryon, or secondary mycelium, contains nuclei of both strains. Heterokaryosis is usually achieved by pairing, either color mutants, or nitrate-nonutilizing (nit) mutants. The lack of complementation of mutant strains after pairing indicates vegetative incompatibility, and it has been demonstrated in several genera of ascomycetes and imperfecti fungi such as *Aspergillus* (Jinks et al. 1966), *Cochliobolus* (Leach and Yoder 1983), *Fusarium* (Puhalla 1985), *Gibberella* (Puhalla and Spieth 1985), *Leptographium* (Zambino and Harrington 1990), *Neurospora* (Perkins 1988), *Podospora* (Fincham et al. 1979), *Sclerotinia* (Kohn et al. 1990), *Septoria* (Newton and Caten 1988), and *Verticillium* (Puhala and Hummel 1983).

Thirty-five strains of *Xylaria cubensis* were tested for mycelial interactions by means of macroscopic examination of pairings on agar medium in all possible combinations and scoring the resulting reaction type. Compatible strains were not tested for heterokaryosis (heterokaryon complementation). The principal objective of undertaking vegetative compatibility studies was to determine intraspecific heterogeneity among endophytic strains, and to estimate genetic diversity within populations of *X. cubensis* from the same and from different leaves.

1.7.3. Study site

The present research was conducted in the floodplains of the Brazilian Amazon at Combu Island (Fig. 1), during January and September of 1989, and 1990. The study site supports a type of vegetation known as tidal "várzea" in the Amazon estuary (Prance 1979), where *E. oleracea* flourishes. It is necessary to make some distinction concerning terminology used for the

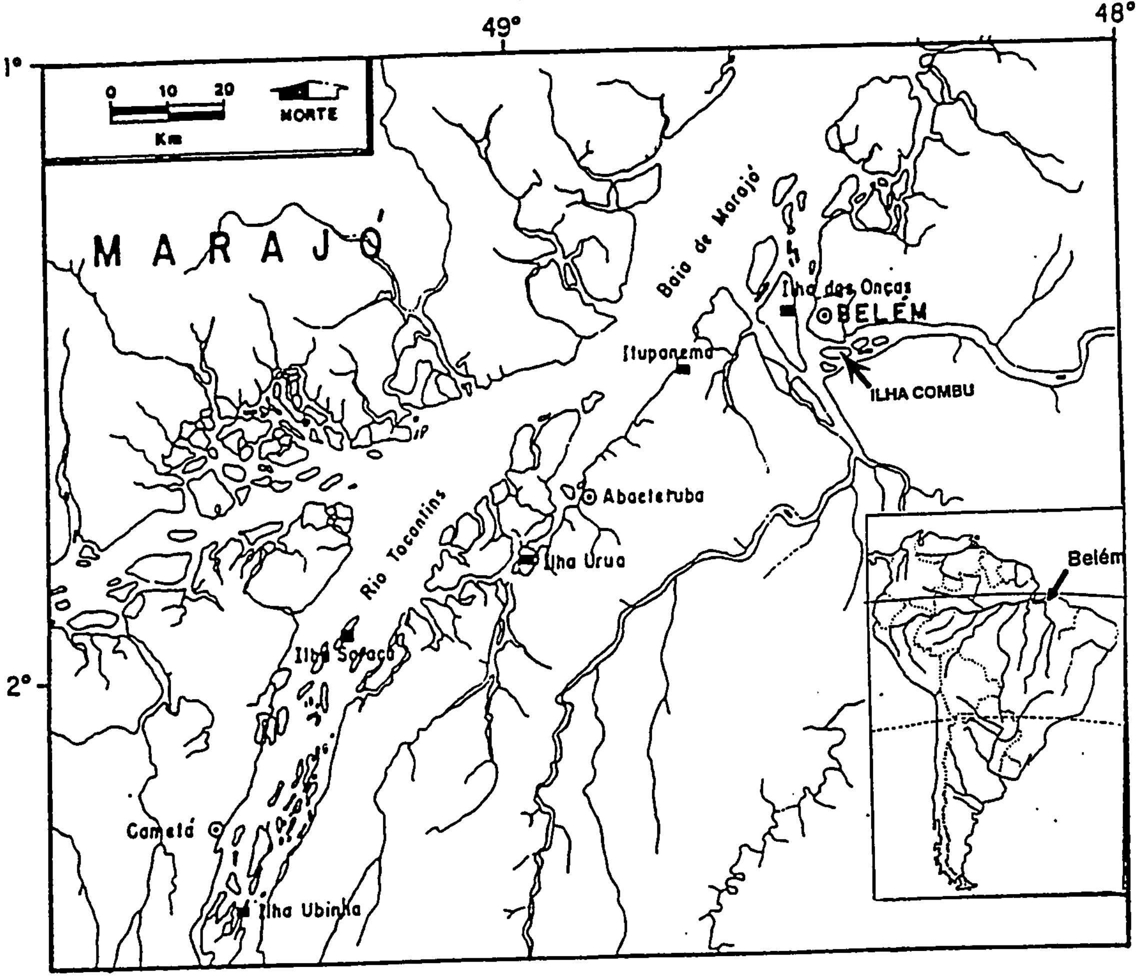


Fig. 1. Geographic location of Combu Island. Courtesy of A. B. Anderson.

Amazonian forests subjected to inundation because of previous confusion. Most of the "white-water" rivers (with a high amount of suspended sediments) have developed areas of floodplains in the middle and lower Amazon, which are called "várzea" (Sioli 1975; Junk 1984), and these represent about 3.8 % of the Amazon Basin (Falesi 1986). In the "tidal várzea", a forest is flooded and drained twice daily by tides at intervals of twelve hours. In "seasonal várzea" flooding is annual due only to the seasonal rise of the white-water rivers from increased rainfall. In the estuarine floodplain of the Combu Island, the strongest influence is from daily tidal flooding. High tides obstruct the rivers flow, with the resultant flooding of nearby land. During the wet season the daily floods are amplified by the greater volume of water in the river resulting from high rainfall.

Combu Island is the site of a management project entitled "Usos tradicionais da terra como alternativa para desenvolvimento do estuário Amazonico" which is being undertaken by the Museu Paraense Emilio Goeldi (MPEG). The main focus of the research is conservation and management of *Euterpe oleracea* Mart. One of the reasons for such a project on this particular island, among dozens of others within the Amazon estuary, is due to its geographical proximity to the city of Belém, the main Amazonian market and place where most fruit of *E. oleracea* is sold commercially (A. Anderson, pers. comm. 1989). This research will complement ecological studies already in progress on the island by providing information that will give a better understanding of the biology of this palm. The project's research station on Combu Island is well situated for carrying out my field work.

1.7.4. Host

Euterpe oleracea was chosen for study because of its ecological and economic importance in the floodplains of the Brazilian Amazon (Calzavara 1972; Anderson 1990a). This palm is known for its ability to recover deep soil nutrients, thus it is widely used in site restoration (Anderson 1988). Every part of the tree is used by people (Fig. 2), making it one of the most valuable plants in the region. The palm hearts and fruits are the two principal uses of açaizeiro as a food source. The former is even exported to other countries, requiring management research in order to prevent over exploitation. Preliminary results of research undertaken by Anderson (1990b) in Combu Island have shown that açaí trees produce about 16 tons of fruits/hectare/year. Therefore, it has been possible for the inhabitants of Combu to rationally exploit açai to give them an annual income of approximately US$ 3,000 per capita; allowing a standard of living that is high for the region (Anderson, 1990b). The management work carried out in Combu consists mainly of selective harvesting of the ramets, where a multiple-stemmed tree is reduced from 12 to 5 stems, i.e., palm hearts are collected only from the ramets, leaving intact the mother plant.

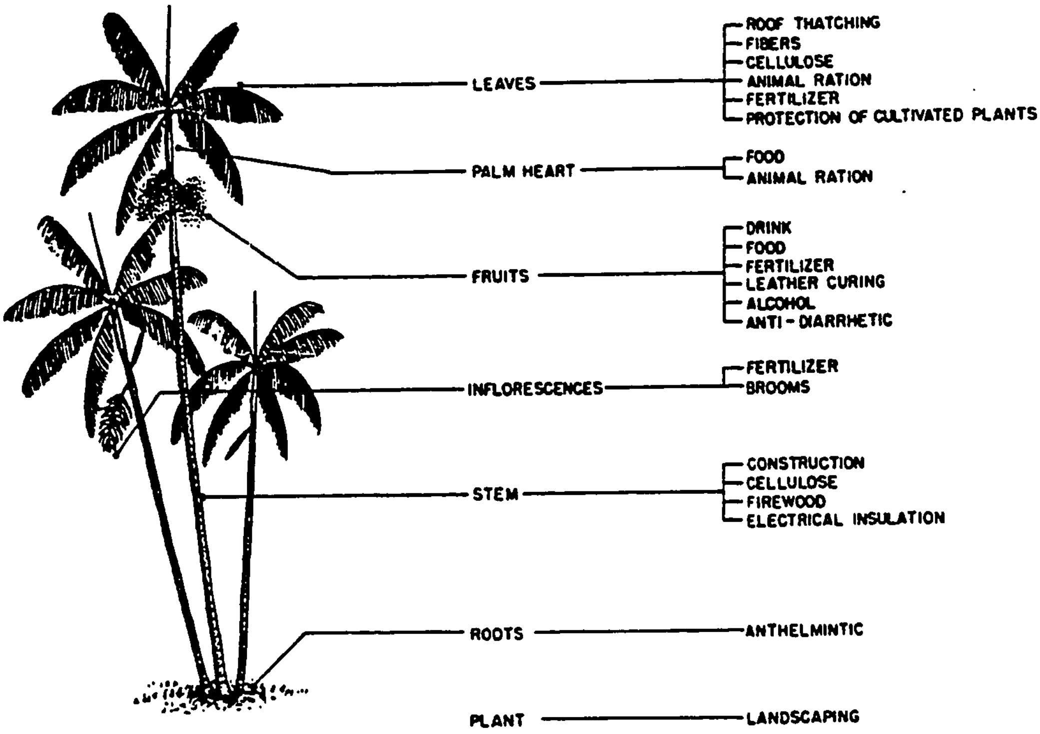


Fig. 2. Economic uses of *Euterpe oleracea*. Courtesy of A. B. Anderson.

2. MATERIALS AND METHODS

2.1. General remarks

Trees and seedlings sampled during this investigation were located inside plots located at two subsites, designated low "várzea" and high "várzea". All trees found within these plots had been previously tagged by other workers as a result of an inventory made in the island. Each tag contained a code formed by six algorithms, e. g. tree # 17 16 02 (located at plot # 17, sub-plot # 16, tree # 02). Trees and seedlings sampled are listed on Tables 1, 2, and 3 with their respective code numbers.

The samples were processed in the phytopathology laboratory at EMBRAPA (Brazilian Agricultural Research Organization) / CPATU (Center of Agricultural Research for the Humid Tropics), in Belém.

2.2. Study site

The study site is a periodically inundated forest located in the floodplains of the Brazilian Amazon at Combu Island in the vicinity of Belém (01°28'S, 48°27'W), state of Pará. The climate is the Afi type, according to the Köppen classification (Köppen 1923). It is characterized by a prominent rainy season, high air relative humidity, and no cool season. The annual rainfall is 2,800 mm, and the average temperature is 26°C . January and February are the wettest months; September and October the driest (driest month with over 60 mm of rain).

Combu Island measures 5 x 3 km (John Rumbold, pers. comm. 1990), and is located in the Guamá River, surrounded by Furo do Paciencia (a channel that communicates with the Guamá River) and Furo do Benedito (Fig. 3). Twenty plots, each 90 x 100 m, are located in the study site (Fig. 3). These are situated either on low várzea (elevation < 1 m), or high várzea (elevation > 1 m). Plot numbers (Tables 1, 2) located at low várzea are 17, 18, 19, 20, and at high várzea are 6, 7, 8, 9, 12. The topographical differences found on várzea forests are due to the accumulation of heavier particles, which are deposited first near river banks, forming the so called "high várzea". The lighter particles are carried away from the river banks and are deposited in the interior of the forest, forming the "low várzea." The forest in the low várzea is adapted to constant inundation. It is dominated by *Euterpe oleracea* Mart., and to a lesser extent by other palms such as *Mauritia flexuosa* L. ("buriti"), and *Raphia taedigera* Mart., an introduced crop *Theobroma cacao* L. ("cacau"), and a monocotyledoneous shrub *Montrichardia arborescens* Schott. ("aninga"), which is typical on river banks of "várzeas". Components of the vegetation at high várzea are *Hevea* spp. ("seringueira"), *Hymenaea courbaril* L. ("jutaí"), *Inga* spp., *Platonia insignis* Mart. ("bacuri"), *Protium krukovii* Swart ("breu"), *Spondias mombin* Urb. ("taperebá"), and *Virola surinamensis* (Rol.) Warb. ("ucuúba"). Although *E. oleracea* is also found in the high várzea, it is rare.

2.3. Host

Euterpe oleracea (Palmae, Arecoideae) is an arborescent, multiple stemmed palm (Fig. 1). Under natural conditions, each *E.oleracea* tree might form up to 12 short stems, which are known as ramets (Anderson et al. 1985).

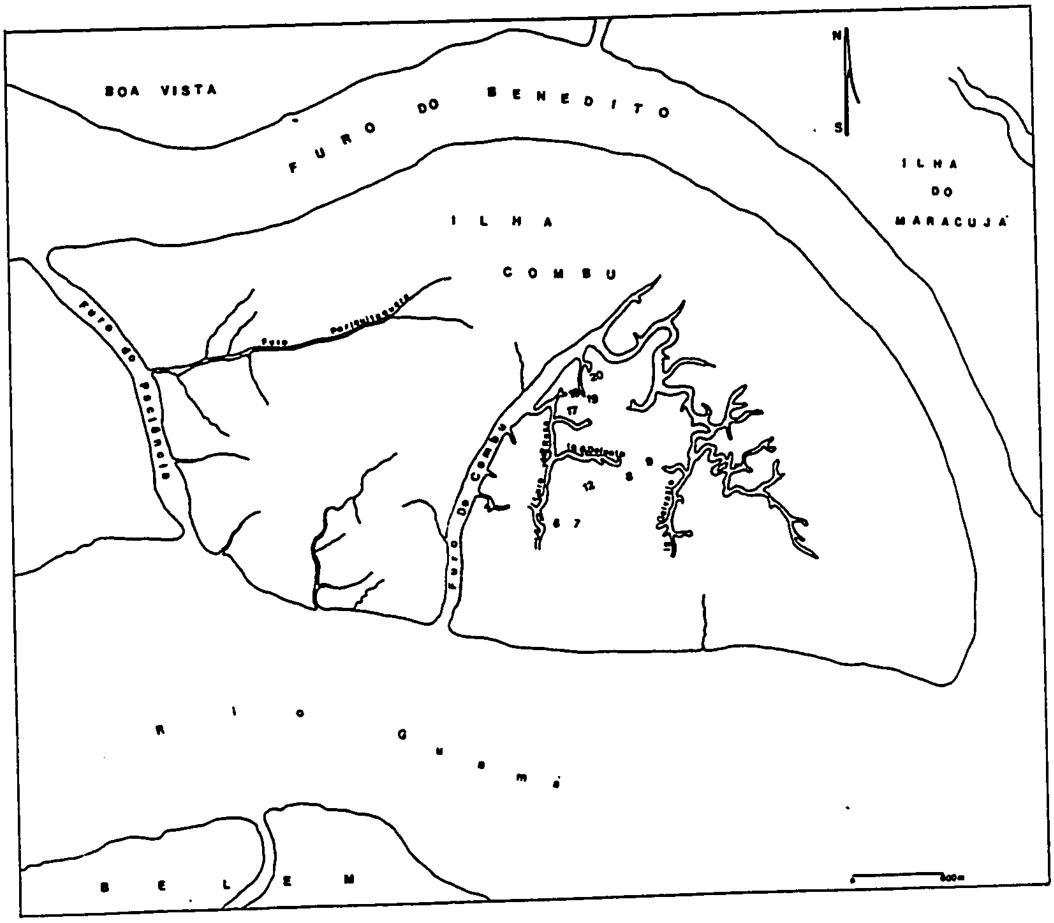


Fig. 3. Plot locations from where *Euterpe oleracea* leaves were collected. Courtesy of the Botany Department, Museu Paraense Emilio Goeldi, Pará state, Brazil.

It has pinnate leaves, and slightly pendulous, subopposite, lanceolate leaflets with a prominent adaxial midrib. This species is a New World palm, ranging from the Amazon estuary, the Orinoco Delta, the Guianas (Guyana, Surinam, French Guyana), and the Pacific coast of Colombia and Ecuador. *Euterpe oleracea,* a native palm in the Amazon, is known locally by its vernacular name - açaizeiro, or açai do Pará. Pollination of *E.oleracea* is primarily by insects belonging to the Hymenoptera, and dispersal of the seeds is generally by birds (Calzavara 1972).

There are no reports of fungal diseases affecting açaizeiros (Calzavara 1972; 1988). The possible reason of that is because açaizeiros are usually not cultivated on large scale. Instead, most of the commercial exploitation of açaí is derived from trees naturally growing along rivers of the Amazon Basin.

2.4. Sampling

All trees and seedlings (listed on Tables 1, 2, 3) were chosen at random, and in four field trips sampled over a two year period, during the wet (January-February 1989/1990) and dry (September-October 1989/1990) seasons.

The age and height of each tree was estimated "in loco" by field technicians.

Trees and seedlings were marked with colored plastic tape when they were first collected in January of 1989 in order to be easily located for the subsequent collecting. With two exceptions, the same trees were continuously sampled. Seedlings could only be sampled once because the

Table 1. Trees (T) investigated during wet and dry seasons, at low, and high várzea.

Tree code	Plot #	Age (years)	Height (m)
Low várzea			
T 1: # 17 16 02	17	15	20
T 2: # 18 20 02	18	6	9
T 3: # 19 11 04	19	7	9
T 4: # 20 03 14	20	20	25
T 5: # 20 19 02	20	7	12
High várzea			
T 6[1]: # 06 05 03	06	5	9
T 6[1]: # 06 18 01	06	7	12
T 7[1]: # 06 12 03	06	5	9
T 7[1]: # 06 20 04	06	7	8
T 8: # 08 19 02	08	5	8
T 9: # 08 17 03	08	5	9
T 10: # 08 18 01	08	5	8

[1] Trees # 6, and # 7 were replaced by # 06 18 01, and # 06 20 04 respectively, during collections made in 1990.

Table 2. Seedlings investigated during wet and dry seasons, at low várzea.

Seedling #	Plot #	Age (months)	Height (m)
January 1989:			
1	17	6	1.5
2	18	6	2.0
3	19	6	2.0
4	20	9	2.5
5	20	9	2.5
September 1989:			
1	17	6	1.5
2	18	6	1.5
3	17	6	1.5
4	20	18	3.5
5	20	9	2.5
January 1990:			
1	17	9	2.5
2	18	6	1.5
3	17	6	1.5
4	20	6	1.5
5	20	6	1.5
September 1990:			
1	17	6	1.5
2	18	9	2.0
3	19	9	1.5
4	20	12	3.0
5	20	12	2.5

Table 3. Seedlings investigated during wet and dry seasons, at high várzea.

Seedling #	Plot #	Age (months)	Height (m)
January 1989:			
6	6	6	2.0
7	6	9	2.5
September 1989:			
6	12	6	1.5
7	6	18	3.0
8	7	5	1.5
9	7	12	2.5
10	12	6	1.5
January 1990:			
6	12	6	2.0
7	8	6	1.5
8	8	6	1.5
9	12	6	2.5
10	12	6	2.5
September 1990:			
6	9	6	1.5
7	8	12	2.5
8	8	6	1.5
9	9	6	1.5
10	6	6	1.5

sampling technique frequently resulted in the introduction of disease or even killed the seedling outright.

Leaves were always collected from the main stem of adult trees. Seedlings were either ramets, or individual seedlings. To avoid the possibility of pathogenic fungi being isolated, only apparently healthy leaflets were chosen.

Trees and seedlings were sampled as follows. From each tree and seedling three fronds belonging to three age classes were collected, viz. one unopened leaf (L1), one newly expanded leaf (L2), and one mature leaf (L3). In January 1989, five trees and five seedlings from the low várzea, and two trees and two seedlings from the high várzea were sampled. In September 1989, January 1990, and September 1990, five trees, and five seedlings, each from low and high várzea were sampled. Thus, a total of 10 trees and 10 seedlings were investigated. Due to intense flooding during January 1989, the high várzea was largely inaccessible and only two trees, and two seedlings could be collected.

2.5. Processing

Three fronds from each tree and seedling were cut into ordered and numbered segments, from base (#1) to tip (#5). Each segment averaged approximately 50 cm in length. Segments were carried in polyethylene bags to the laboratory at EMBRAPA where they remained inside closed bags in the laboratory at room temperature (25°C). Samples were processed within 24 hours of collecting.

Whole leaflets (20 per frond) were surface sterilized by consecutive submersion for 1 min in 75% ethanol, 10 min in 65% domestic bleach ("Brillux", 5% sodium hypochlorite), and 30 sec in 75% ethanol. Thirty discs of approximately 3 mm diameter from the midribs (herein called veins), and 30 from the interveins (space between veins) of the leaf blade of each frond (180 discs per tree) were cut with a flame-sterilized corkborer. Thus, a total of 13,320 discs were taken for the four sampling periods (2,520 discs for the first sampling, and 3,600 discs for each of the reamining three samplings). Five discs were evenly distributed in plastic Petri dishes (9 cm diam.) containing cornmeal dextrose agar (CMD, Difco), supplemented with 4 g/l Streptomycin sulfate (Appendix 1) to inhibit bacterial growth, and 5 mg/l of Cyclosporine A (CsA, Sandoz; Appendix 1) to prevent fast growing fungi from masking slow growing species (Dreyfuss 1986). In addition, CsA allowed more time to process the expected large number of colonies. The Petri dishes were sealed with Parafilm, incubated in the laboratory under conditions of alternating diffused daylight and darkness at approximately 25°C, and examined periodically over a period of 30 days.

New colonies appeared continuously from 5 to 30 days. Each colony was isolated onto a culture tube containing cornmeal agar (CM, Difco) as it appeared. Each isolate received a code number indicating its origin. Notes were immediately taken on general colony characteristics. Culture tubes were transported to the New York Botanical Garden for identification and further studies.

2.6. CULTURAL WORK

The standard medium used was CMD. Cultures were grown in 9 cm plastic Petri dishes, and incubated at 20°C under a cycle of 12 hours fluorescent light and 12 hours darkness. Growth rates were calculated after 5 to 7 days of incubation, and expressed as diameter measurements.

Data on gross colony morphology was routinely recorded, e.g. growth rate, color, texture, margin, zonation, pigment diffusion. In addition, data on morphology of reproductive structures such as sexual (ascocarp, basidiocarp) and asexual organs (conidiophores, pycnidium, synnema, stroma) were also taken. Microscopic characteristics were taken from observation of freshly mounted material in water, Melzer's reagent (for staining ascus tips, showing a positive reaction by turning blue: J^+), cotton blue in lactic acid (for staining fungal protoplasts), or Amann's lactophenol (for semi-permanent mounts). An Olympus model BH-2 compound microscope equipped for brightfield and phase contrast (BH2-PC) microscopy was used. Microscopic features, such as conidial morphology, were recorded using a drawing tube (BH2-DA).

Color designations used throughout the taxonomic descriptions are from Kornerup and Wanscher (1978), and appear inside parenthesis following the color names.

The terminology used herein to describe texture of mycelia followed Stalpers (1978), and morphological structures followed Ellis (1971), Hawksworth et al. (1983), Kendrick (1971), and Sutton (1980).

Representative cultures derived from this research are deposited in the American Type Culture Collection (ATCC). Selected isolates are maintained on CM agar and are stored at 8°C at the New York Botanical Garden.

2.7. *Xylaria* species

2.7.1. Isozyme analysis

Eighty-one isolates of *Xylaria* species were used in the isozyme analysis, and are listed by the stock, reference numbers, and source, in Table 4.

Preparation of isolates for the isozyme work was done at the Systematic Botany and Mycology Laboratory of the USDA, (Beltsville, MD) as follows.

Five mycelial plugs were taken from the edge of actively growing, young fungal colonies growing on CMD in Petri dishes by using a flame sterilized cork borer (11 mm diam), and then placed into sterile Eberbach blender bowls containing 100 ml CYM liquid medium (Appendix 1). The plugs and medium were blended for 30 seconds at low speed in a Waring blender. Fifty ml of blended medium were poured into each of two 125 ml flasks. Flasks were placed on a rotary shaker at 150 rpm at 20°C in darkness. The isolates were incubated for 10 to 14 days, at which time there was abundant mycelium production.

2.7.2. Harvest

Mycelial mats were collected on No. 1 Whatman filter paper in a Büchner vacuum funnel. Each mat was washed with 50 ml distilled water.

Table 4. Isolates of *Xylaria* species used in the isozyme analysis.

Stock #	Reference #	Strain #	Source	Site
X. adscendens				
91-05	22	301v1e	T7L3	H
91-54	7	289v2d	T10L2	H
91-81	31	301i2a	T7L3	H
91-384	41	444i5b	S8L2	H
91-409	34 39[1]	445v1e2	S8L3	H
X. allantoidea				
89-16	52	75v3a	S3L3	L
91-182	2	310i1a	S8L3	H
91-229	15	334v6d	S5L1	L
91-327	66	480i2a	T6L3	H
91-441	14	108v1c	S4L2	L
91-442	1	60i1a	T7L3	H
X. anisopleura				
89-12	56	41v1c	T2L3	L
91-282	137	296v2c	T6L3	H
91-594	138	109v3a	S4L2	L
X. arbuscula				
91-91	82	300v1e	T7L3	H
91-177	78	340i1b	S6L2	H
91-183	83	303i1d	T7L2	H
91-247	85	347v5a	S9L2	H
91-249	86	335v2b	S2L3	L
91-275	88	322i3b	S1L3	L
91-306	70	436v4c	T10L2	H
91-426	55	445i4a	S8L3	H
91-428	77	483i1a	T7L3	H
X. castorea				
89-134	4	90v3d	S4L2	L
89-150	48	1i2a	T1L3	L
89-152	47	1v6a	T1L3	L
91-331	53	465v1e	T9L3	H
91-524	65	421v3b	S3L3	L
X. coccophora				
91-171	58	303i2b	T7L2	H
91-174	59	302v2e	T7L3	H
91-260	33	305v2b	T7L2	H
91-540	75	436v6a1	S10L2	H

Table 4. (cont.)

Stock #	Reference #	Strain #	Source	Site
X. cubensis				
91-105	20	334vld	S5L1	L
91-200	23	347i6d	S9L2	H
91-374	3	458ilc	T10L3	H
91-390	8	441vc	S6L2	H
91-410	29	449v2d	T8L2	H
91-533	63	436v2d	S10L2	H
X. curta				
91-124	71	314vld	S7L2	H
91-125	64	331i4d	S10L2	H
91-184	84	283v2b	T8L2	H
91-280	49	336i3a	S2L2	L
91-301	50	441v5c	S6L2	H
91-303	13	444i6d1	S8L2	H
91-305	6	456i2b	T10L3	H
91-333	68	442i5a	S6L3	H
91-406	72	444i3b	S8L2	H
91-411	74	459ve	T10L3	H
91-412	87	444v6e	S8L2	H
91-413	73	466vla	T9L3	H
91-414	62	433ilc	S7L2	H
91-420	35	433i2b	S7L2	H
X. ? microceras				
89-27	11	71v4a	S3L2	L
91-398	10	395vb	T1L3	L
91-427	40	428vlb	S1L3	L
X. multiplex				
89-116	17	133vla	T4L3	L
90-98	24	98vld	S7L3	H
91-93	80	286ila	T8L3	H
91-109	79	293vlal	T6L2	H
91-123	76	335v5a	S2L3	L
91-176	21	344i4a	S4L3	L
91-235	43 38[1]	322i3a	S1L3	L
X. obovata				
91-408	19	426vla	S1L2	L
91-465	36	415v3b	S4L2	L
91-466	32	364vlb	T2L3	L
91-469	42	438v2a	S9L2	H
91-504	67	415v2a	S4L2	L

Table 4. (cont.)

Stock #	Reference #	Strain #	Source	Site
X. ? palmicola				
91-88	12	297i1a	T6L3	H
91-340	5	485v1c	T7L3	H
X. ? telfairii				
89-119	30	198v1a	T6L2	H
89-157	133	198v1e	T6L2	H
90-192	18	304i2c	T7L2	H
91-107	60	295v2d	T6L2	H
91-142	27	334v5a	S5L1	L
91-153	25	291v4c	T10L3	H
91-310	26	418i4a	S2L2	L
	9 [1]			
	16 [1]			
	46 [1]			

Source: T=tree, S=seedling, L=leaf. Site: H=high, L=low.

[1] Isolates derived from teleomorphs cultured in the laboratory of Prof. Jack D. Rogers (Washington State University, WSU):
X. adscendens:# 39: Hawaii, on *Sapindus saponaria*.
X. multiplex: # 38: SM 1491, Mexico, on unident. dicot.
X. telfairii: # 9, 16, 46: 7708-0307 NTU, Taiwan, on unident. dicot.

Harvested mycelia were placed in plastic weighing boats, and stored in a freezer at -60°C.

2.7.3. Lyophilization

Frozen mycelia were transferred to plastic centrifuge tubes and covered with cheesecloth. Up to 12 tubes were placed in a stainless steel beaker, attached to a multi-dry/freeze-dryer (FTS Systems, Inc.), and lyophilized at -60°C under 80 millitorr vacuum for approximately 18 hours. The dry mycelia were then transferred to sealed plastic bags and kept at -60°C for three weeks.

2.7.4. Sample Preparation

Samples were processed for gel electrophoresis in the Laboratory of Plant Systematics (Geobotany Department) at Eidgnossische Technische Hochschule (ETH), Zürich, as follows. Mycelia were ground to a fine powder using a mortar and pestle, then placed in glass vials (60 x 16.0/1.0 mm - GEWINDE 137/5 - AR-KLARGLAS). The labeled vials were kept at -35°C. Twenty-five mg of mycelia were suspended in 0.3 ml of 0.1 M Tris-HCl-PVP extraction buffer (pH 7.5) (Appendix 2), and kept in Eppendorf tubes at 4°C overnight. Samples were centrifuged at 14,000 rpm for 15 minutes (Sigma 201 M Centrifuge). The pH adjustments were performed by using Mikroprozessor pH meter 761 - Knick (Elektronische Messgeräte) Berlin, Germany.

2.7.5. Starch-gel

Starch gels were prepared by suspending 57.6 g (12.8 %) hydrolyzed starch (Sigma) in 450 ml of gel buffer in a 1 liter Erlenmeyer flask. The mixture was heated over a Bunsen burner (with continuous swirling) until it started boiling. The suspension was then degassed with a vacuum formed by a water aspirator, to the point where all small bubbles were removed from the solution. The gel was poured into an acrylic gel tray, covered with a glass plate to avoid desiccation, and left overnight at room temperature to solidify.

2.7.6. Loading samples and electrophoresis

Prior to applying samples, the gel was placed in a refrigerator for about one hour.

A slit was cut across the length of the gel about 4 cm from the cathodal edge of the gel. A ruler (marked with 32 dots corresponding to the total number of samples) was placed on top of the gel, very close to the slit, and centered. The Eppendorf tubes were removed from the centrifuge and placed into a bowl containing ice. The samples were loaded as follows. The contents of each Eppendorf tube were absorbed onto filter paper wicks (2 x 13 mm, 0.8 mm thick; Whatman) held by forceps (# 2). Each wick was quickly blotted on paper towels, and completely inserted into the slit. Up to 32 samples were loaded on each gel.

Two filter paper wicks contained a dye marker (0.04% bromophenol blue) and were applied to either side of the samples.

The gel tray was then put inside an electrode tray containing electrode buffer, and run at 4° C. The power supply routinely used was: Electrophoresis Power Supply EPS 500/400 - Pharmacia LKB Biotechnology (Sweden).

Three buffer systems were used: **I**: electrode buffer citric acid, pH 7.0; gel buffer histidine.HCl, pH 7.0; 75mA constant current until marker dye migrated 6 cm. **II**: electrode buffer Tris citric acid, pH 8.0; gel buffer Tris citric acid, pH 8.0; 50mA constant current until marker dye migrated 9 cm. **III**: electrode buffer Tris citric acid, pH 7.2; gel buffer Tris citric acid, pH 7.2; 50mA constant current until marker dye migrated 9 cm.

After 10 to 15 minutes all filter paper wicks were removed. A ruler was inserted at the cathodal edge (between gel and tray) to prevent separation of the two slices of gel, when the wicks were removed.

2.7.7. Enzyme staining

Enzyme staining solutions were prepared according to the protocols outlined by Soltis et al. (1983). Recipes of buffers, and enzyme stains are given in the Appendix 2. Enzyme names with respective enzyme commission (EC) number (I.U.B. 1984), abbreviations, and buffer systems used for this work are given in Table 5.

On completion of electrophoresis, the gel was sliced (up to six per gel) horizontally using an adaptation of a saw with a steel string. Each slice was put into boxes of approximately 10 x 16 cm for enzyme staining. The staining reaction was stopped when bands were well defined, at which time the staining solution was poured off, and the gel was washed with tap water.

Initially 21 enzyme stains were tested for activity with the three buffer systems. Five enzymes were selected which showed clear bands in most of the isolates examined. The following enzymes were examined but not included here, due to their streaking bands or inconsistent activity: acid phosphatase (E.C. 3.1.3.2), aconitase (E.C. 4.2.1.3), alcohol dehydrogenase (1.1.1.1), alkaline phosphatase (3.1.3.1), aspartate aminotransferase (2.6.1.1), ß-D-glucosidase (3.2.1.21), diaphorase (1.6.4.3), fructose-bisphosphatase (3.1.3.11), fumarase (4.2.1.2), glucose-6-phosphate dehydrogenase (1.1.1.49); hexokinase (2.7.1.1), isocitrate dehydrogenase (1.1.1.42), leucine aminopeptidase (3.4.11.1), malate dehydrogenase (1.1.1.37), malic enzyme (1.1.1.40), shikimate dehydrogenase (1.1.1.25).

2.7.8. Interpretation

Gel slices were examined under ultraviolet light (Kaiser prolite 5000) to interpret band patterns and to make migration distance measurements. The gels were photographed using Technical Pan film (Kodak).

The electromorphs assigned for each enzyme were determined by repeated side by side comparisons and expressed as values that reflected their increasing mobility on the gel, i.e., from the most anodal to the most cathodal edge.

Since the genetic basis of the electromorphs studied is not known, the isozyme data were interpreted phenotypically, i.e. each band pattern was treated as a different electromorph. This procedure follows studies previously done by Leuchtmann and Clay (1990), and Leuchtmann et al. (1992).

Table 5. Enzyme names with enzyme commission (EC) number, abbreviations, and buffer systems used with *Xylaria* species.

Enzyme	EC No.	Abbreviation	Buffer system
Aldolase	4.1.2.13	**ALD**	II
Phosphoglucose isomerase	5.3.1.9	**PGI**	III
Phosphoglucomutase	5.4.2.2.	**PGM**	III
6-phosphogluconate dehydrogenase	1.1.1.44	**6PG**	I
Triosephosphate isomerase	5.3.1.1	**TPI**	III

2.8. *Xylaria cubensis* - complex:

2.8.1. Isozyme analysis

Isozyme analyses were performed with 35 isolates as listed in Table 6. Sample preparation followed the protocol routinely used in the Microbiology Institute at ETH, Zürich. The inoculum was grown on 2% malt extract agar (MEA) in Petri dishes. Three 5 mm diam discs were taken from the edge of actively growing colonies, and placed in 50 ml liquid V8 medium (Appendix 1) in 100 ml Erlenmeyer flasks (one flask for each isolate). The flasks were incubated in the dark, on a rotary shaker (110 rpm) at 20°C. Mycelia were harvested after 17 days and lyophilized by using a vaccuum pump Lyovac GT 2 (Leybold - Heraeus GMBH, Köln-Germany). Enzyme names with enzyme commission (EC) number, abreviations, and buffer systems used are given in Table 7. Of 12 enzyme systems examined for activity, six were selected because they gave clear bands. Aldolase (4.1.2.13), aspartate aminotransferase (2.6.1.1), diaphorase (1.6.4.3), and isocitrate dehydrogenase (1.1.1.41) were not included because of the production of streaking bands. Acid phosphatase (E.C. 3.1.3.2) and hexokinase (E.C. 2.7.1.1) were excluded because they did not show activity at all.

Table 6. Isolates of *Xylaria cubensis* used for the isozyme and Vegetative Compatibility Group analyses.

Reference #	Stock #	VCG Reference #
3	91-374	27
8	91-390	35
23	91-105	33
29	91-410	1
101	91-603	7
102	91-612	19
103	91-618	34
104	91-616	13
105	91-608	23
106	91-611	14
107	91-600	16
108	91-602	30
109	91-32	29
110	91-33	21
112	91-610	6
113	91-103	24
114	91-620	5
115	91-623	25
116	91-605	31
117	91-604	28
118	91-24	2
119	91-597	26
120	91-617	18
121	91-598	11
122	91-607	3
123	91-621	10
124	91-609	17
125	91-599	8
126	91-606	9
127	91-615	4
128	91-601	12
129	91-622	22
130	90-236	20
131	91-613	15
132	91-619	32

Table 7. Enzyme names with enzyme commission (EC) number, abbreviations, and buffer systems used with *Xylaria cubensis*.

Enzyme	EC No.	Abbreviation	Buffer system
Aconitase	4.2.1.3	**ACO**	I
Malate dehydrogenase	1.1.1.37	**MDH**	III
Phosphoglucose isomerase	5.3.1.9	**PGI**	III
Phosphoglucomutase	5.4.2.2.	**PGM**	III
6-phosphogluconate dehydrogenase	1.1.1.44	**6PG**	I
Triosephosphate isomerase	5.3.1.1	**TPI**	III

2.8.2. Numerical analysis

A set of 11 characters for each isolate, including cultural morphology and data from the isozyme analyses, were chosen for the numerical taxonomy (Sneath and Sokal 1973). Taxonomic character state coding was either binary (for presence or absence of features) or multistate coding (e.g., for isozyme phenotypes) and is presented in Table 8.

2.8.3. Vegetative Compatibility Group test (VCG)

Thirty-five isolates of *Xylaria cubensis* (Table 9) were crossed in all possible combinations on 2% Malt extract agar (MEA, Difco).

The crosses were prepared as follows. Plugs 3 mm diam were cut with a flame-sterilized cork borer from the margin of actively growing colonies, and placed on 9 cm plastic Petri dishes. Three plugs, each from a different isolate, were spaced 1.5 cm from each other near the center of plates. Because self-crosses were also accomplished, two plugs of the same isolate were placed 1.5 cm apart in separated plates. Cultures were incubated for one week in the dark, at room temperature (20°C), and then examined for compatibility, i. e. either intermingling or the formation of reaction lines (dark brown to blackish pigmented lines) between isolates. The reactions were scored using the following criteria: **1** for somatically compatible strains (intermingling), and **0** for somatically incompatible strains (formation of a light to heavily pigmented line).

Table 8. Characters and character states coded for *Xylaria cubensis*-complex.

1	**Isozyme phenotype**	A: 1, B: 2, C: 3, D: 4, E: 5, F: 6, G: 7, H: 8, I: 9, J: 10, K: 11, L: 12, M: 13, N: 14, O: 15.
	Colony:	
2	color	Pale salmon: 1, pale salmon then darkening to brownish: 2.
3	growth rate (in 7 days)	3.5 cm diam: 1, 4 cm: 2, 4.5 cm: 3, 5 cm: 4.
4	texture	Cottony: 1, cottony with radial strands: 2, appressed with a fast sector: 3.
5	zonation	No: 0, yes: 1.
6	margin	Even: 1, lobed: 2.
7	reverse	Non-pigmented: 0, pigmented: 1
	Stroma	
8	shape	Cylindrical: 1, flabelliform: 2 both present: 3
9	distribution	Central: 1, peripheral: 2, all over: 3.
10	color	Pale salmon: 1, pale salmon, darkening to brown at base: 2.
11	conidia size	3.6-4.5 x 1.8-2.7 µm 5.4-6.3 x 2.7-3.6 µm

Table 9. Isolates of *Xylaria cubensis* used in the VCG test.

Reference #	Stock #	Source	Plot #	Site
1	91-410	T8L2	8	H
2	91-24	S4L2	20	L
3	91-607	T8L2	8	H
4	91-615	S8L2	8	H
5	91-620	S8L3	8	H
6	91-610	T8L3	8	H
7	91-603	S8L2	8	H
8	91-599	S8L3	8	H
9	91-606	S8L2	8	H
10	91-621	S8L3	8	H
11	91-598	S10L3	12	H
12	91-601	S10L2	12	H
13	91-616	S8L2	8	H
14	91-611	T8L3	8	H
15	91-613	T8L3	8	H
16	91-600	S10L2	12	H
17	91-609	T8L3	8	H
18	91-617	S10L2	12	H
19	91-612	S10L2	12	H
20	90-236	S5L1	20	L
21	91-33	S4L2	20	L
22	91-622	T8L2	8	H
23	91-608	T8L3	8	H
24	91-103	S10L3	12	H
25	91-623	T8L2	8	H
26	91-597	S8L2	8	H
27	91-374	T10L3	8	H
28	91-604	S10L3	12	H
29	91-32	S10L3	12	H
30	91-602	T8L2	8	H
31	91-605	S8L3	8	H
32	91-619	S10L3	12	H
33	91-105	S5L1	20	L
34	91-618	S10L2	12	H
35	91-390	S8L3	8	H

Source: L=leaf, S=seedling, T=tree. Example: T8L2 (tree # 8, leaf 2). Site: H=high, L=low.

2.9. Statistical methods

2.9.1. Endophytic communities

A data base (d-BaseIII Plus) for each field trip was created in order to organize the data. A data matrix was prepared using a spreadsheet software (Lotus 1-2-3, release 2.3) to be used with the statistical software package SYSTAT 5.0 (Wilkinson 1989).

The percentage of colonization per tree was calculated in the following manner: 180 discs that were taken from each tree (60 discs from leaf 1, 60 from leaf 2, and 60 from leaf 3) at each sampling period was considered to be 100%. In the case of colonization rate per leaf, the percentage was calculated by considering the value 60 as 100%.

A *G*-test (Sokal and Rohlf 1981) of goodness of fit was performed to test whether the overall colonization rates for the four sampling periods were statistically different.

Differences in colonization frequencies due to the different levels of factors such as plant age, season, and subsite were assessed by one-way ANOVA (analysis of variance) using a general linear model (GLM), because of the unbalanced nature of the model.

Two techniques of community ordination study, Principal Component Analysis (PCA) and Nonmetric Multidimensional Scaling (MDS), were performed based on the rate of infection presented by the twenty-one most frequently isolated fungi. The statistical analyses were undertaken on a reduced matrix containing only the most frequent species (at least 1% relative importance of one species). A Minimum Spanning Tree was drawn where the distance between every pair of points of a set of line segments was linked by only one path (Pielou 1984). The combinations of letters and numbers used in the MDS graphs encode the sampling units location (L=low site, H= high site), plant developmental stage (T= tree, S= seedling), plant tissue (i= intervein, v= vein), and sampling periods (1, 2, 3, 4= January 1989, September 1989, January 1990, September 1990, respectively).

2.9.2. Isozyme analysis

A community classification technique, Cluster Analysis (CA), was applied to the resulting electromorphic matrix containing all isozyme phenotypes. A normalized percent of disagreement distance was used in order to measure dissimilarities between two individual clusters. A dendrogram resulted from the clustering techniques where the intercluster distances were computed by means of average-linkage clustering (unweighted pair-group mean average - UPGMA) [Sneath and Sokal 1973].

2.9.3. Numerical analysis

A PCA was carried out on a data matrix (Appendix 5) containing the character states coding of the *Xylaria cubensis*-complex in order to determine which characters were the most significant, i.e., characters that would affect the OTUs (Operational Taxonomic Units - isolates in this case). A normalized percent of disagreement was computed for each pair of isolates, then analyzed by means of average-linkage cluster procedures.

3. RESULTS

3.1. Overall colonization and endophytic communities

From a total of 13,320 leaf discs incubated during four sampling periods, 3,200 fungal isolates were obtained. Fifty-seven species and six familial taxa were recognized. Table 10 shows all identified fungi and the total number of times that each was isolated during the collecting periods.

Euterpe oleracea trees and seedlings showed a low incidence of colonization by endophytic fungi. Overall colonization rates (defined as the total number of infected discs divided by the total number of discs incubated, expressed in percentages) both for trees and seedlings, of 30.3 %, 23 %, 21 %, and 24 %, respectively for the first, second, third, and fourth sampling periods were obtained. The *G*-test indicated differences with regard to overall colonization rates significant at the 0.01 level ($G = 5{,}996.53 > X^2\ 0.01[3] = 11.34$).

The distribution of endophytic fungi was highly affected by the leaf age, plant developmental stage and subsite. The ANOVA test (Appendix 7) showed that differences detected in the colonization frequencies from leaves of different ages ($P < 0.001$), from trees and seedlings ($P < 0.001$) and from low and high várzea ($P < 0.001$) were statistically significant. In relation to the season, the pattern of endophyte distribution was not significant at the 0.05 level, i.e., colonization frequencies were relatively constant during the wet and dry seasons.

Table 10. Overall colonization frequencies of endophytic fungi isolated from *Euterpe oleracea* during four evaluation periods. I=Jan 89, II=Sep 89, III=Jan 90, IV=Sep 90, T=total.

Taxon	I	II	III	IV	T
Acremonium sp.	7	2	1	0	10
Acrodictys elaeidicola	6	4	1	15	26
Anthostomella sp.	10	4	2	0	16
Arthroconidial	15	17	7	6	45
Basidiomycetes	2	3	3	1	9
Calonectria sp.	0	0	0	1	1
Chloridium ? preussii	1	0	2	1	4
Colletotrichum gloeosporioides	29	17	8	4	58
Colletotrichum sp.	0	0	3	7	10
Curvularia pallescens	4	5	1	0	10
Daldinia eschscholzii Anamorph: *Nodulisporium*	0	2	0	0	2
Dendrodochium sp.	6	0	0	0	6
Fusarium oxysporum	0	2	1	3	6
F. sacchari v.*elongatum*	6	0	0	0	6
F. semitectum v. *majus*	1	0	2	0	3
F. verticillioides	4	3	6	2	15
Graphium sp.	1	0	0	0	1
H. quisquiliarum Anamorph: *Nodulisporium*	1	5	1	2	9
Hypoxylon serpens Anamorph: *Geniculosporium*	15	53	12	67	147

Table 10. (cont.)

Taxon	I	II	III	IV	T
Hypoxylon sp. Anamorph: *Virgariella*	4	0	1	0	5
Hypoxylon stygium Anamorph: *Nodulisporium*	18	14	25	49	106
Idriella amazonica	3	3	0	0	6
I. asaicola	1	2	0	3	6
I. euterpes	32	11	5	13	61
Lasiodiplodia theobromae	2	5	1	0	8
Leiosphaerella cocoes	4	3	0	1	8
Letendraea sp.	122	211	50	100	483
Microdochium sp.	0	2	0	1	3
Mycelia Sterilia	57	37	32	13	139
Mycoleptodiscus sp.	2	0	0	0	2
Neosartoria sp.	3	0	0	0	3
Nigrospora sphaerica	2	0	0	0	2
Nodulisporium sp.	2	7	7	3	19
Oxydothis poliothea	1	0	0	2	3
Penzigia indica Anamorph: *Nodulisporium*	1	0	3	0	4
Pestalotiopsis palmarum	10	13	0	0	23
Phoma sp.	17	8	6	6	37
Phomatospora sp.	5	6	3	9	23
Phomopsis sp. 1	35	17	16	29	97
Phomopsis sp. 2	6	0	1	0	7

Table 10. (cont.)

Taxon	I	II	III	IV	T
Physalacria sp.	1	0	0	0	1
Physalospora sp.	2	0	0	1	3
Thozetella sp.	20	39	22	44	125
Trichoderma sp.	2	0	0	0	2
Ustulina ? deusta Anamorph: *Nodulisporium*	0	26	6	4	36
Wardomyces sp.	3	8	1	0	12
Xylaria adscendens Anamorph: *Nodulisporium*	26	17	59	13	115
X. allantoidea Anamorph: *Nodulisporium*	4	5	2	4	15
X. anisopleura Anamorph: *Nodulisporium*	6	5	2	5	18
X. arbuscula Anamorph: *Nodulisporium*	10	13	94	42	159
X. castorea	4	0	3	3	10
X. coccophora Anamorph: *Nodulisporium*	3	2	6	4	15
X. cubensis complex Anamorph: *Xylocoremium*	131	156	209	177	673
X. curta Anamorph: *Nodulisporium*	1	6	8	107	122
X. ? microceras	4	0	0	3	7
X. ? multiplex	14	5	27	14	60
X. obovata Anamorph: *Nodulisporium*	0	1	0	8	9
X. ? palmicola	0	0	1	1	2

Table 10. (cont.)

Taxon	I	II	III	IV	T
X. ? telfairii	4	8	12	1	25
Xylariaceae I	12	24	18	40	94
Xylariaceae II	24	18	21	8	71
Xylariaceae III	17	24	28	25	94
Xylariaceae IV	25	14	9	7	55
Xylariaceae V	15	12	19	2	48
Total	763	839	747	851	3200

The total number of isolates for individual trees and seedlings of *E. oleracea* for each sampling time is presented in Figs. 4, 5, 6, and 7 based on the total number of discs infected.

The colonization rates (CR) in trees varied between 9% and 46% for the first sampling period, 2% and 29% for the second, 2% and 40% for the third, and 2% and 36% for the fourth sampling period. In seedlings the CR varied from 14% to 51% during the first sampling period, 20% and 50% during the second, 13% and 34% during the third, 23% and 53% during the fourth sampling period. Tree # 4 showed the lowest CR during all sampling periods, and this was a pattern shared among other trees located at low várzea, while the majority of trees located at high várzea showed the highest CR.

3.1.1. Discussion of selected endophytic fungi

The fungi most commonly found in leaves of *E. oleracea* were the ascomycetes *Xylaria cubensis* (21% of the total number of discs infected) and *Letendraea* sp. (15% of the total number of discs infected).

The endophytic fungal community in leaves of *E. oleracea* presented distinct patterns of species composition and frequency. Three groups of endophytes could be distinguished. The first was characterized by two species abundantly present throughout all sampling times (*X. cubensis*, and *Letendraea* sp.), the second by species consistently isolated but at lower frequency (representing 48.3% of the total number of discs infected), and the third, the remaining species

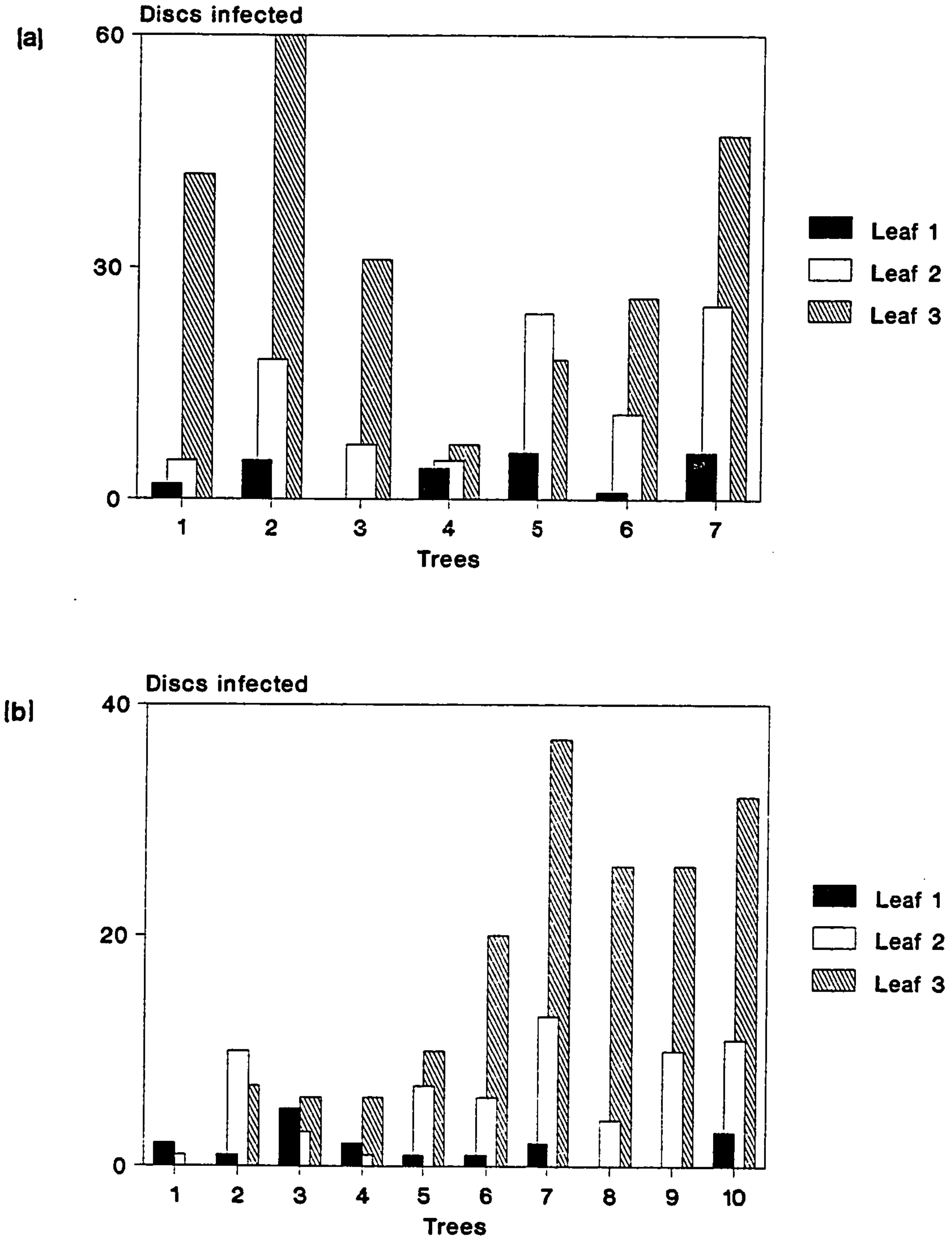


Fig. 4. Total number of isolates per tree and leaf age. Sampling periods: (a) Jan. 1989, and (b) Sep. 1989. Leaf 1: unopened leaf, leaf 2: newly expanded, leaf 3: mature leaf. Trees 1 to 5: located at low várzea, trees 6 to 10: high várzea.

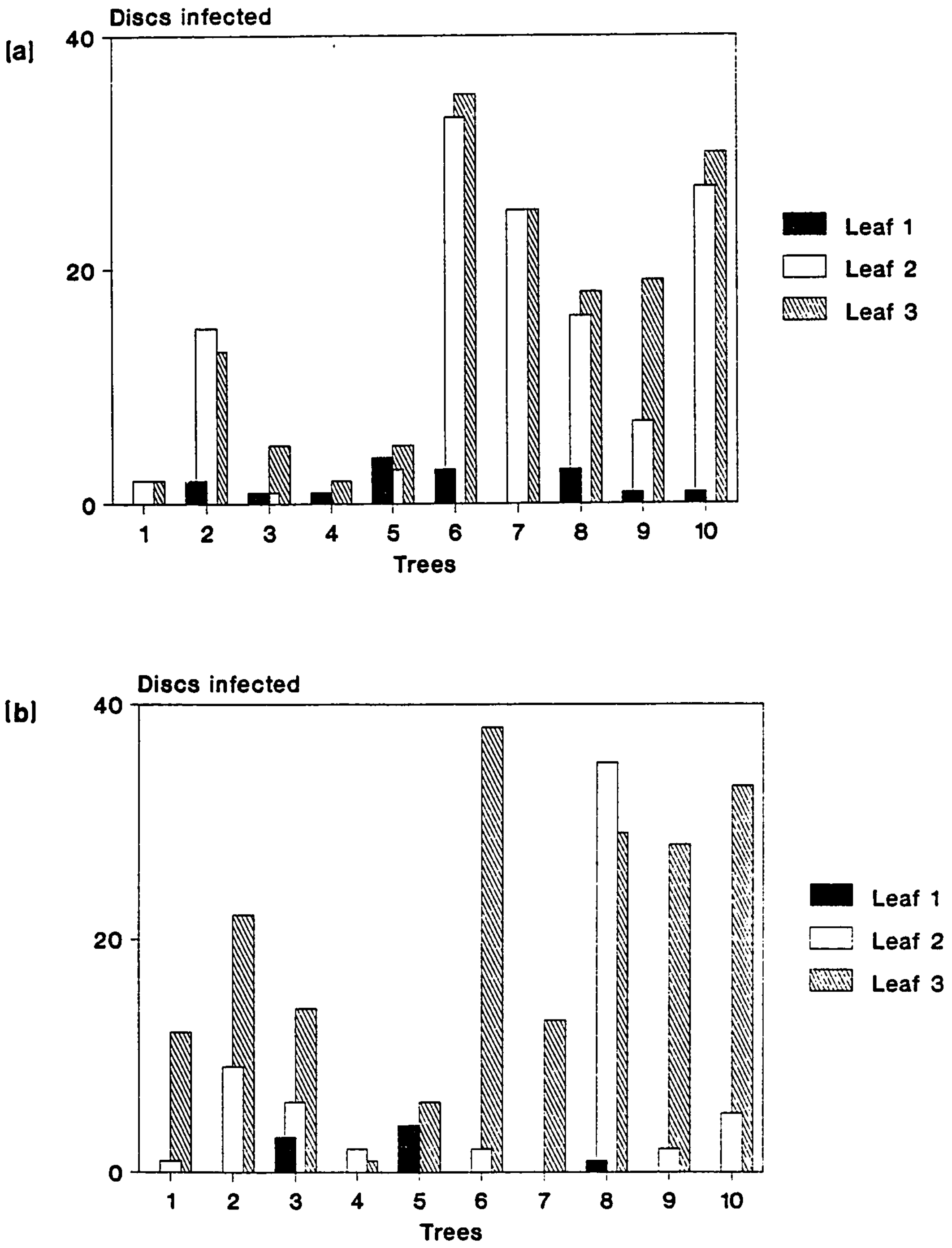
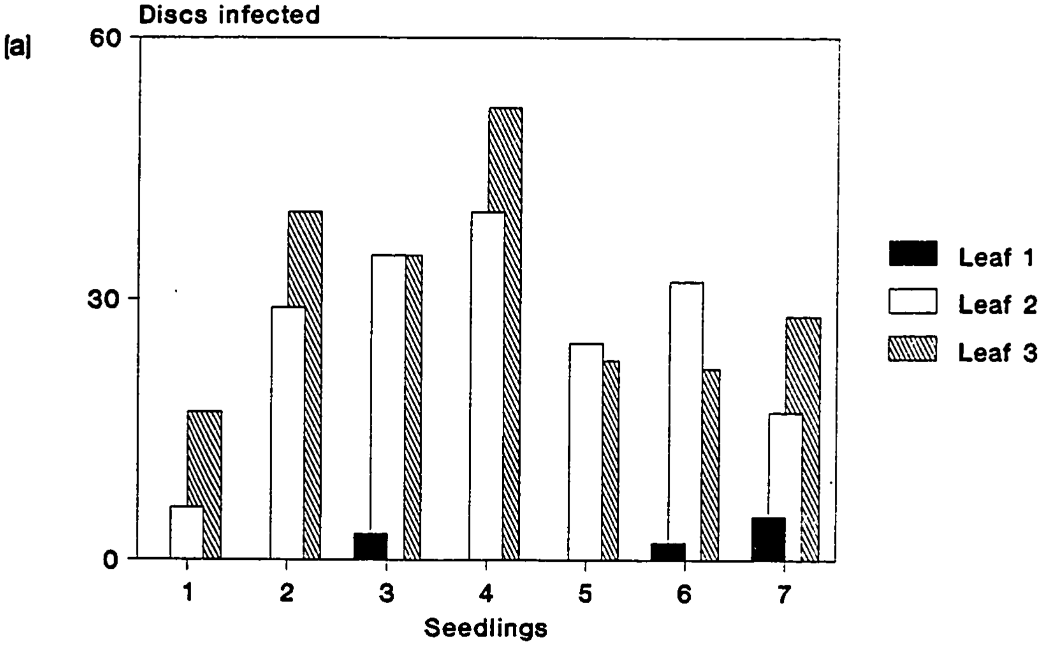


Fig. 5. Total number of isolates per tree and leaf age. Sampling periods: (a) Jan. 1990, and (b) Sep. 1990. Leaf 1: unopened leaf, leaf 2: newly expanded, leaf 3: mature leaf. Trees 1 to 5: located at low várzea, trees 6 to 10: high várzea.



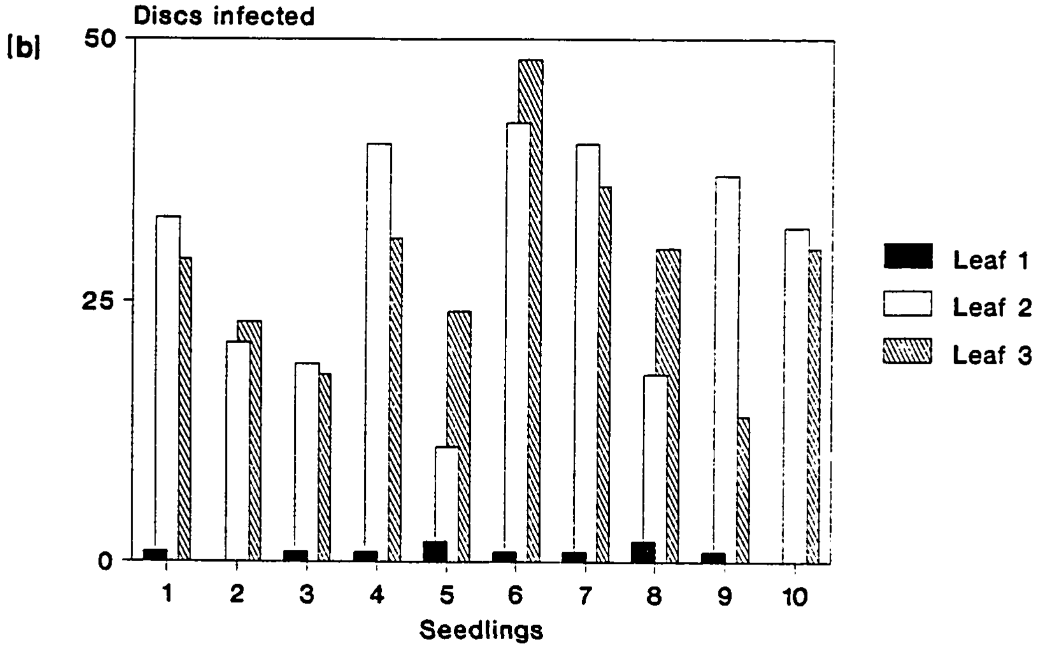


Fig. 6. Total number of isolates per seedling and leaf age. Sampling periods: (a) Jan. 1989, and (b) Sep. 1989. Leaf 1: unopened leaf, leaf 2: newly expanded, leaf 3: mature leaf. Seedlings 1 to 5: located at low várzea, seedlings 6 to 10: high várzea.

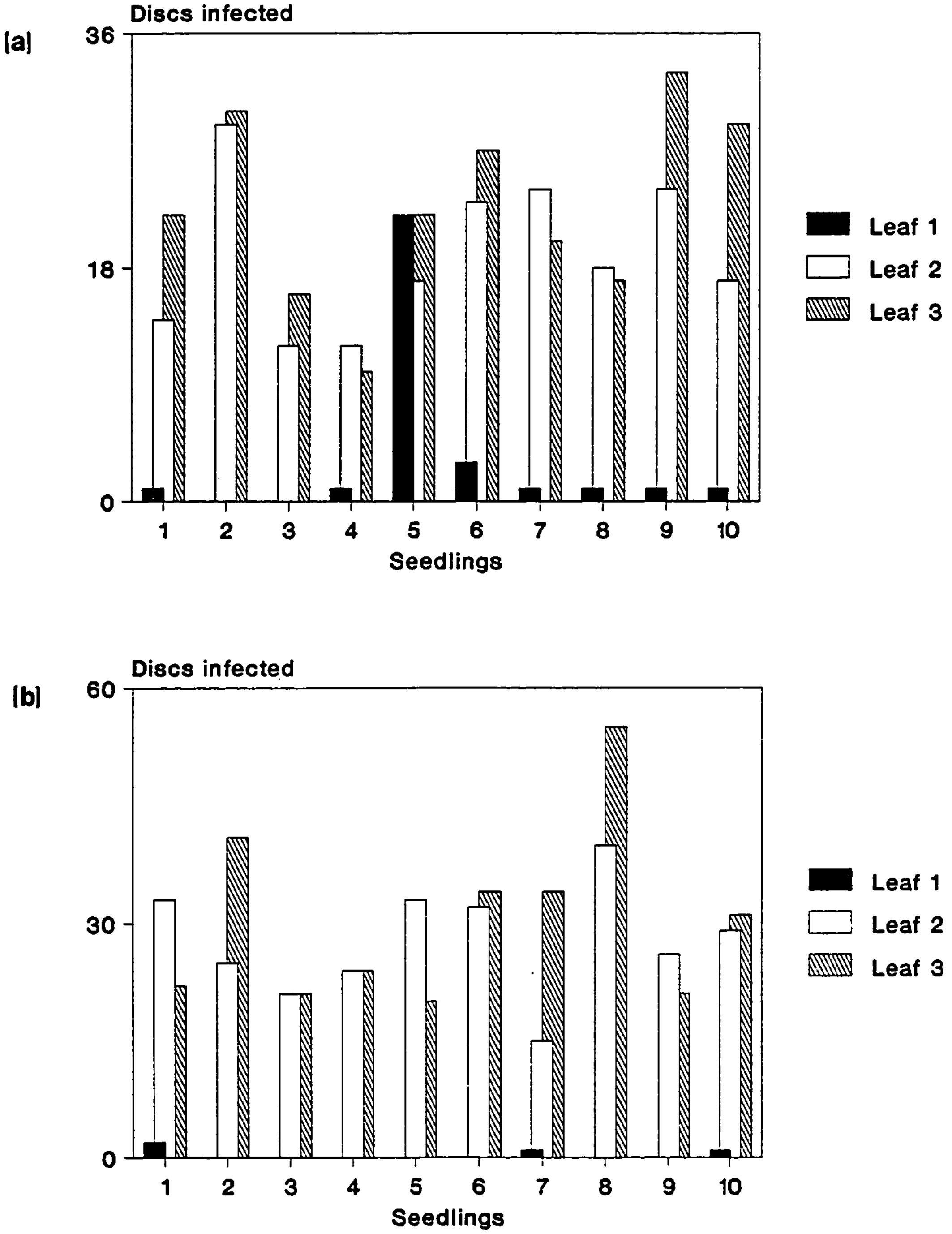


Fig. 7. Total number of isolates per seedling and leaf age. Sampling periods: (a) Jan. 1990, and (b) Sep. 1990. Leaf 1: unopened leaf, leaf 2: newly expanded, leaf 3: mature leaf. Seedlings 1 to 5: located at low várzea, seedlings 6 to 10: high várzea.

that were sporadically or rarely isolated, representing 15.7% (of the total number of discs infected). The species that fall into the second group, in descending order, are *Xylaria arbuscula*, *Hypoxylon serpens*, *Thozetella*, *X. curta*, *X. adscendens*, *H. stygium*, *Phomopsis* sp. 1, Xylariaceae I, Xylariaceae III, Xylariaceae II, *Idriella euterpes*, *X. multiplex*, *Colletotrichum gloeosporioides*, Xylariaceae IV, Xylariaceae V, Arthroconidial fungus, *Phoma*, *Ustulina deusta*, and *Acrodictys elaeidicola.*

Differences in the occurrence of endophytes were more pronounced in relation to the leaf tissue location (vein vs. intervein), plant developmental stage (tree vs. seedling), and sampling subsite (high vs. low várzea) rather than between the collecting periods (dry/wet season). These aspects are discussed in more detail in the following paragraphs.

3.1.2. Leaf age effects

Table 11 shows the overall frequencies of colonization of the twenty-one most frequent fungi according to their occurrence within the three leaf age types (unopened: age 1, newly expanded: age 2, and mature: age 3). In general, most of the fungi were present in all leaf classes. However, changes in colonization frequencies were characterized by increasing numbers of species (Xylariaceae I, III, and IV) as well as increasing in the frequency of species already established with increasing leaf age.

Table 11. Frequency of colonization (actual numbers) per **leaf age** in four sampling periods, of the twenty-one most frequent fungi.
Number of discs sampled per leaf-age = 4,440.

Taxon	**age 1**	**age 2**	**age 3**
A. elaeidicola	1	11	14
Arthroconidial	9	12	24
C. gloeosporioides	1	21	36
H. serpens	2	61	84
H. stygium	4	42	60
I. euterpes	1	21	39
Letendraea sp.	5	200	278
Phoma sp.	12	10	15
Phomopsis sp. 1	14	35	48
Thozetella sp.	1	53	71
U. deusta	1	20	15
X. adscendens	2	48	65
X. arbuscula	6	44	109
X. cubensis	11	249	413
X. curta	1	57	64
X. multiplex	1	18	41
Xylariaceae I	0	45	49
Xylariaceae II	2	32	37
Xylariaceae III	0	43	51
Xylariaceae IV	0	24	31
Xylariaceae V	2	24	22

3.1.3. Leaf tissue-type effects

As suggested by Table 12, endophytes were found to be present in both tissues, vein and intervein. However, preference of one tissue over the other was often observed. For instance, xylariaceous fungi were found in larger numbers in the veins, in contrast to *Letendraea*, *Idriella euterpes*, and the arthroconidial fungi, which were more frequent in the intervein. Other examples of preference for host tissue location were *Colletotrichum gloeosporioides* and *Phomopsis* sp. 1, which were much more frequent within the veins.

Two techniques of community ordination analysis, Principal Component Analysis (PCA) and Nonmetric Multidimensional Scaling (MDS), were performed in order to test the hypothesis that trees and seedlings, located on either low and high várzea could be grouped (clustered) on the basis of the rates of infection by the endophytic fungi. Although PCA allowed some conclusions to be drawn about the endophytic communities of *E. oleracea*, the "horseshoe" disposition of the samples on the graphs suggested non-linearity of the data. Therefore the use of this method was abandoned and MDS was taken up instead. Kruskal's stress (Kruskal, 1964) was used for the scaling. The results of the MDS analyses are presented in Figs. 8, 9, and 10, and they basically confirmed the clusters shown by the PCA analysis.

The analysis showed that sampling sites (low and high várzea) were characterized by distinct endophyte communities. Figures 8, 9, and 10 show the MDS analysis performed for trees and seedlings located in the low and high várzea. These analyses were based on the number of isolates of the

Table 12. Distribution of endophytic fungi in the leaf tissue, during four evaluation periods.
I=Jan 89; **II**=Sep 89; **III**=Jan 90; **IV**=Sep 90; **T**=total.

	VEIN					INTERVEIN				
Taxon	I	II	III	IV	**T**	I	II	III	IV	**T**
A. elaeidicola	3	3	1	11	**18**	3	1	0	4	**8**
Arthroconidial	6	5	2	3	**16**	9	12	5	3	**29**
C. gloesporioides	23	11	7	1	**42**	6	6	1	3	**16**
H. serpens	10	37	7	42	**96**	5	16	5	25	**51**
H. stygium	10	10	15	36	**71**	8	4	11	1	**36**
I. euterpes	7	7	5	3	**22**	25	4	0	13	**39**
Letendraea sp.	22	39	3	10	**74**	99	172	46	89	**406**
Phoma sp.	9	7	3	5	**24**	8	1	3	1	**13**
Phomopsis sp. 1	32	15	14	21	**82**	3	2	2	8	**15**
Thozetella sp.	9	16	12	24	**61**	11	23	10	20	**64**
U. deusta	0	21	5	4	**30**	0	5	1	0	**6**
X. adscendens	21	17	51	10	**99**	5	0	8	3	**16**
X. arbuscula	9	8	78	27	**122**	1	5	16	15	**37**
X. cubensis	90	127	146	125	**488**	41	28	62	51	**182**
X. curta	1	5	5	64	**75**	0	1	3	43	**47**
X. multiplex	9	4	15	9	**37**	5	1	12	5	**23**
Xylariaceae I	4	19	15	24	**62**	8	5	3	16	**32**
Xylariaceae II	19	17	16	7	**59**	5	1	5	1	**12**
Xylariaceae III	11	17	18	17	**63**	6	7	10	8	**31**
Xylariaceae IV	20	10	7	7	**44**	5	4	2	0	**11**
Xylariaceae V	10	8	15	2	**35**	5	4	4	0	**13**

twenty-one most frequently encountered endophytic fungi according to their occurrence within the host tissue (intervein, vein, or both).

There was a general tendency for trees at the two subsites to have the same fungi (Fig. 8,B) and for seedlings in the low site to have a distinct population of endophytes (Fig. 8,A). All trees and most of the seedlings from high várzea formed a cluster (Fig. 8,B). A second less compact group was formed by seedlings located at low várzea (Fig. 8,A).

When fungi found in the intervein region were considered (Fig. 9), the species isolated from trees and seedlings found at high várzea were different from the endophytes isolated from trees in low várzea. In fig. 9, two groups are clearly seen, in the first (Fig. 9,B), all trees and most of the seedlings from high várzea were clustered, indicating a somewhat homogeneous pattern in the group. In the second (Fig. 9,A), all seedlings from the low várzea were clustered. The distances between seedlings from the low várzea (second group - A) are shorter than those between trees and seedlings from the high várzea suggesting a higher degree of relatedness in the second group.

There was a general tendency for the same fungi to be isolated from the vein tissues of trees and seedlings regardless of the site, although the population at high várzea was more homogeneous than the population at low várzea (Fig. 10). A single cluster (Fig. 10,A) comprised all trees and seedlings from the high, and most of the trees from the low várzea.

The low stress of final configuration obtained after Kruskal for the above three analysis were 0.04958, 0.03721, and 0.04354, respectively, indicating the closeness of fit of the model to the data set.

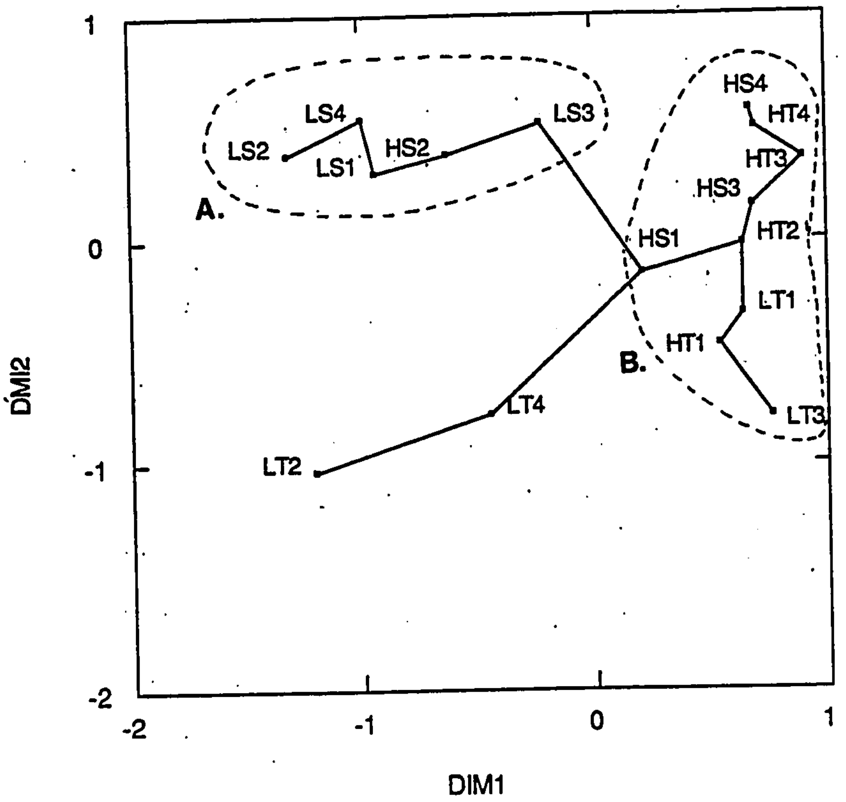


Fig. 8. Results of the Multidimensional Scaling applied to the fungal colonization frequencies. Coefficient of alienation after scaling: 0.04354. L: low várzea, H: high várzea, T: tree, S: seedling, 1-4: sampling periods (Jan. 1989, Sep. 1989, Jan. 1990, Sep. 1990, respectively). See text for explanation.

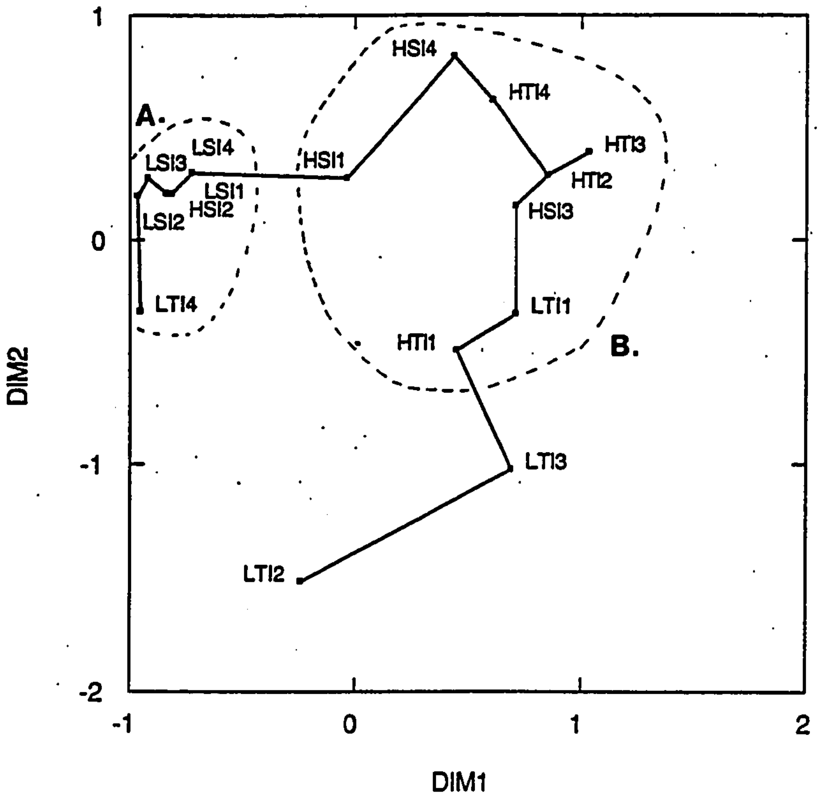


Fig. 9. Results of the Multidimensional Scaling applied to the fungal colonization frequencies in the intervein region. Coefficient of alienation after scaling: 0.04958. L: low várzea, H: high várzea, T: tree, S: seedling, 1-4: sampling periods (Jan. 1989, Sep. 1989, Jan. 1990, Sep. 1990, respectively). See text for explanation.

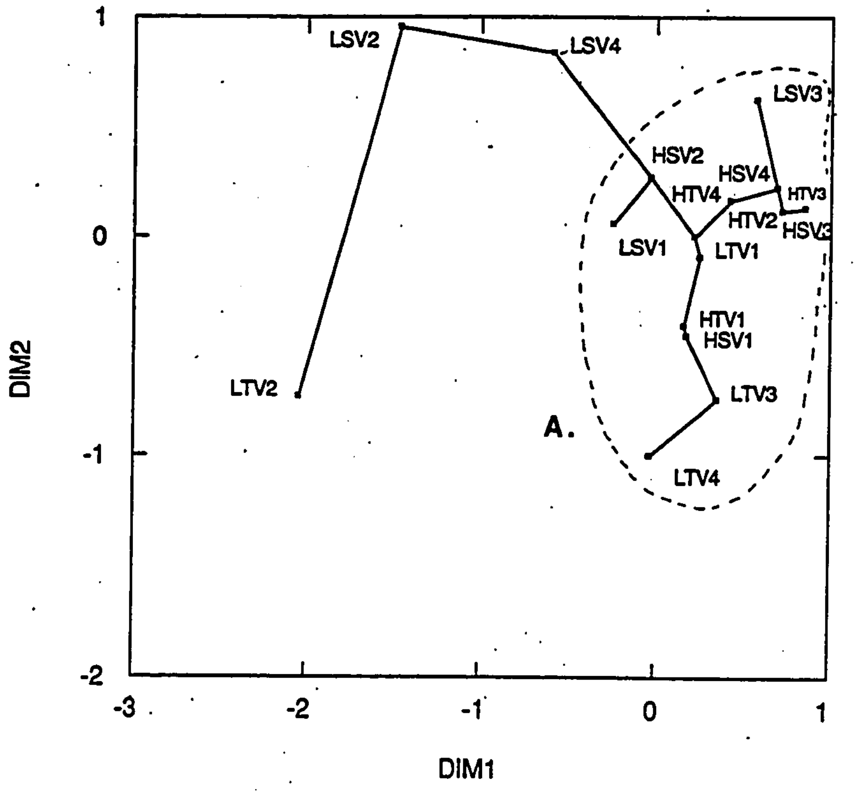


Fig. 10. Results of the Multidimensional Scaling applied to the fungal colonization frequencies in the vein region. Coefficient of alienation after scaling: 0.03721. L: low várzea, H: high várzea, T: tree, S: seedling, 1-4: sampling periods (Jan. 1989, Sep. 1989, Jan. 1990, Sep. 1990, respectively). See text for explanation.

3.1.4. Tree age, and subsite effects

The endophytic communities in seedlings and trees of *E. oleracea* were generally similar but differences were detected in relation to their quantitative composition, revealing some degree of fungal specificity for tree age and subsite (Figs. 11 a,b, 12 a,b, and 13, Appendices 3, 4). As previously mentioned, variation in the occurrence of certain species was noticed among subsites. For instance, *Acrodictys elaeidicola*, *Idriella euterpes*, *Letendraea* sp., and *Thozetella* sp. occurred more frequently in the low várzea. Conversely, most of the species of the Xylariaceae were more frequent in the high várzea.

Figures 11 and 12 show the distribution of the eight most common endophytic species in trees and seedlings during the first, second, third and fourth sampling periods, respectively (Fig. 11a,b, Fig. 12 a,b,), as well as the composition of the fungal communities at high and low várzea during the four sampling periods (Fig. 13).

The total number of isolations of *X. cubensis* was nearly equal both in trees and seedlings during the first two sampling periods. In the last two samplings, there were twice as many *X. cubensis* found in seedlings as in trees. *X. cubensis* was generally more common in the high várzea, than in low várzea, representing 57%, 45%, 50%, and 36% respectively for the first, second, third, and fourth periods (Fig. 13).

Letendraea sp. appeared to colonize differentially tree ages and subsites. In general, *Letendraea* isolates were considerably more frequent in seedlings than in trees throughout the four sampling periods, although a slight decrease was noticed during the last two sampling periods (Figs. 11, 12). This coincides with an increase of *X. cubensis*. It is possible that the increase in frequency of one species is responsible for the decrease of the other, suggesting a possible antagonistic effect between these two species. The percentage of isolations by *Letendraea* in the low várzea was remarkably high, representing 43%, 63%, 22%, and 36%, respectively, for the first, second, third and fourth sampling periods (Fig. 13).

Xylaria arbuscula showed a somewhat stronger preference for seedlings (Figs. 11, 12) than for trees. Its occurrence was relatively low during the first two sampling periods, considerably higher in the third, and declined in the fourth sampling. *X. arbuscula* was generally found in the high várzea, with exception of the third sampling when it was equally present, i.e. ca. 20% in both subsites.

Hypoxylon serpens and *H. stygium* were generally isolated in larger numbers from seedlings (Figs. 11, 12) than from trees. Their percentage of distribution in the subsites differed only slightly from each other. *H. serpens* was more abundant in the high várzea, except for the last sampling (Fig. 13). The percentages of distribution of *H. serpens* in the high várzea were 5%, 14%, and 3%, respectively, for the first, second, and third sampling periods. *H. stygium* seemed to be better represented in the low várzea.

Thozetella was another example of a species highly dependent on tree age and subsite. The occurrence of *Thozetella* in seedlings located at low várzea was remarkable (Fig. 13). The percentages in the low várzea were 7%, 17%, 8%, and 16%, respectively, for the first, second, third, and fourth sampling periods.

The number of isolations of *X. curta* during the first three sampling periods was generally low. This species preferentially colonized seedlings at high várzea, where it represented 26% (Fig. 13).

Xylaria adscendens was isolated most frequently from trees during the first and third sampling periods, and from seedlings during the second and fourth periods. It was better represented in the high várzea, accounting for 10%, 4%, 14%, and 3%, respectively, of the isolates in the first, second, third, and fourth sampling periods.

The distribution pattern of other fungi in trees, seedlings and subsites deserves some attention. For instance, the distribution of arthroconidial fungi suggested a preference for trees, and they were isolated in greater numbers from the low várzea. *Idriella euterpes* displayed a higher isolation frequency at low várzea, but showed no apparent preference for tree age. *Ustulina deusta* was more frequent in seedlings, and was more common at high várzea than at low. *Colletotrichum gloeosporioides* showed no conspicuous specificity either for tree age or subsite.

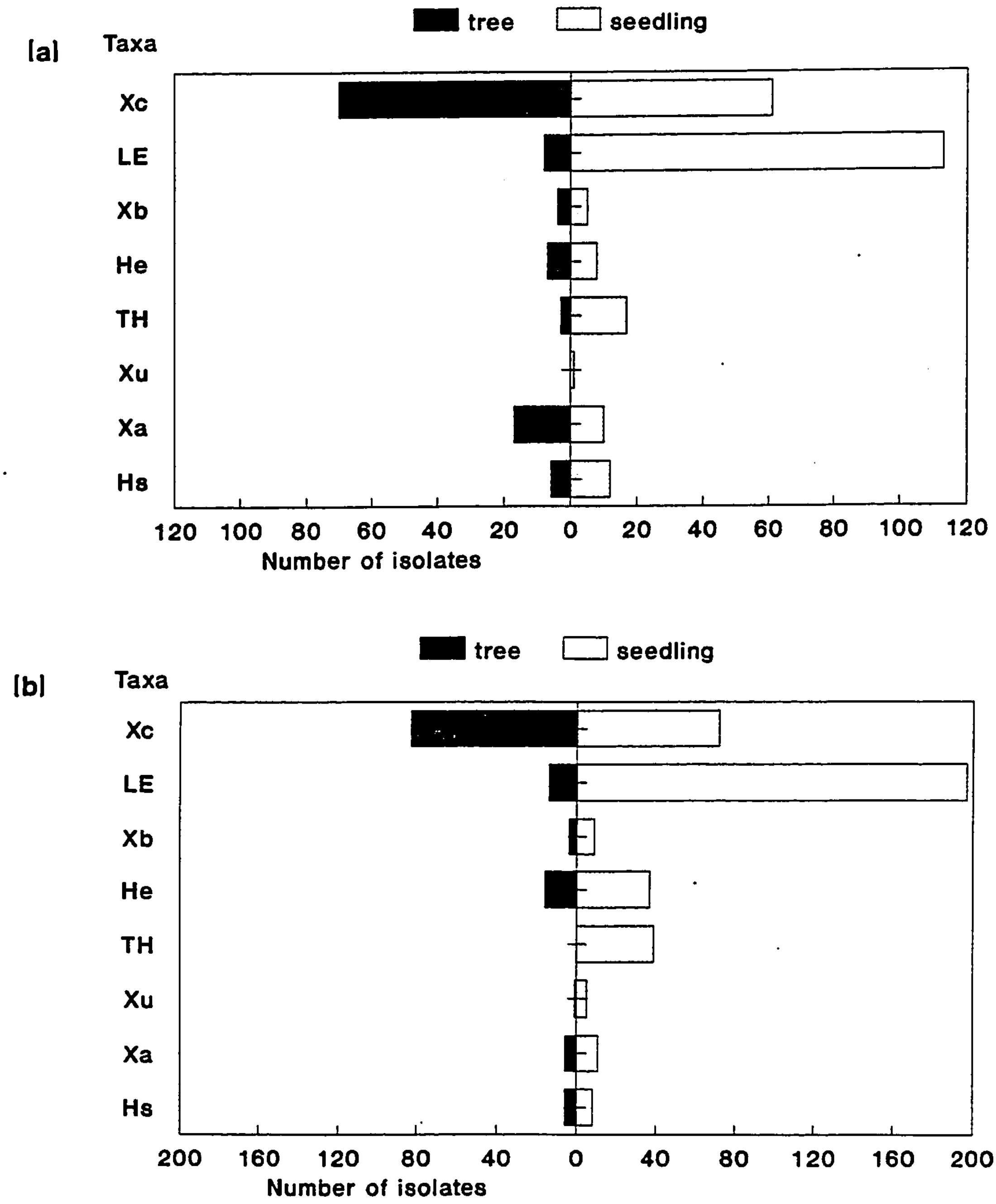
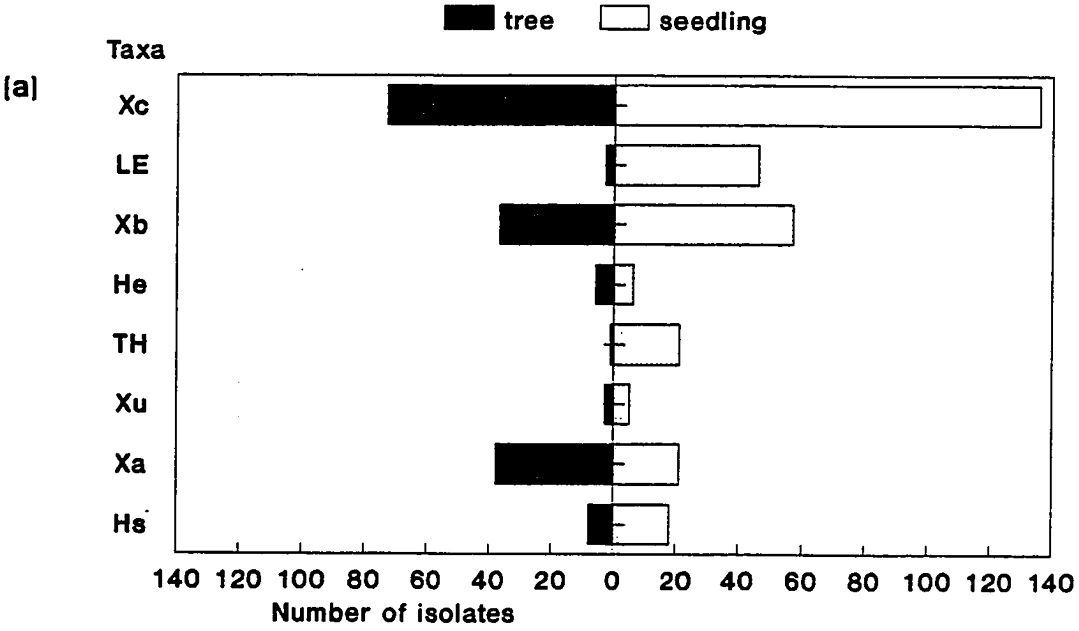


Fig. 11. Distribution of the eight most frequent endophytes in trees and seedlings, in the sampling periods of (a) Jan. 1989, and (b) Sep. 1989.

Xc *Xylaria cubensis*	TH *Thozetella* sp.
LE *Letendraea* sp.	Xu *Xylaria curta*
Xb *Xylaria arbuscula*	Xa *Xylaria adscendens*
He *Hypoxylon serpens*	Hs *Hypoxylon stygium*



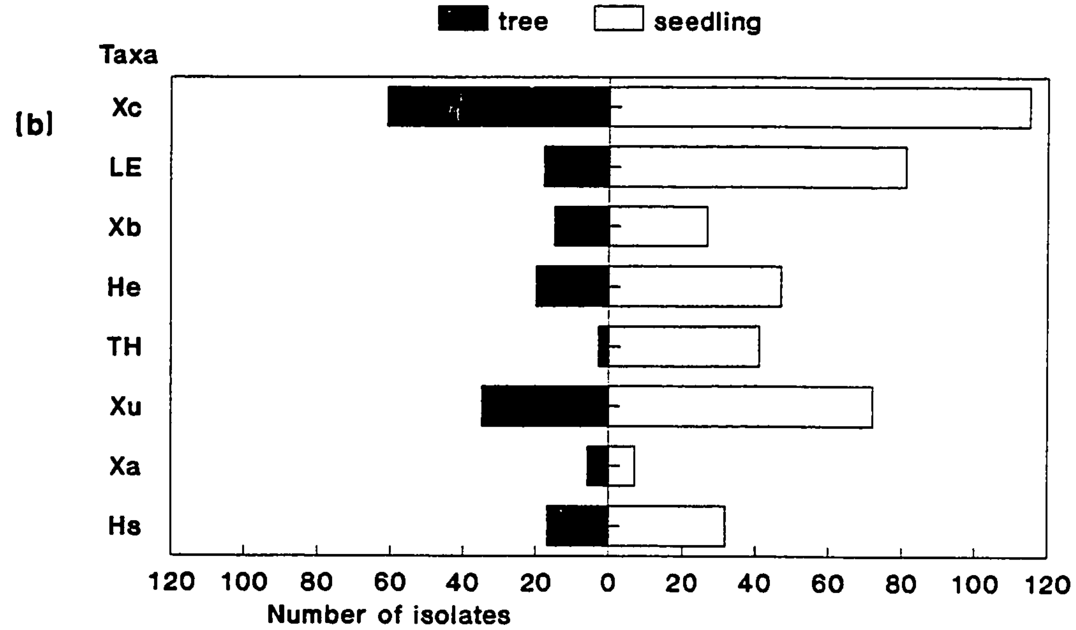


Fig. 12. Distribution of the eight most frequent endophytes in trees and seedlings, in the sampling periods of (a) Jan. 1990, and (b) Sep. 1990.

Xc *Xylaria cubensis*	TH *Thozetella* sp.
LE *Letendraea* sp.	Xu *Xylaria curta*
Xb *Xylaria arbuscula*	Xa *Xylaria adscendens*
He *Hypoxylon serpens*	Hs *Hypoxylon stygium*

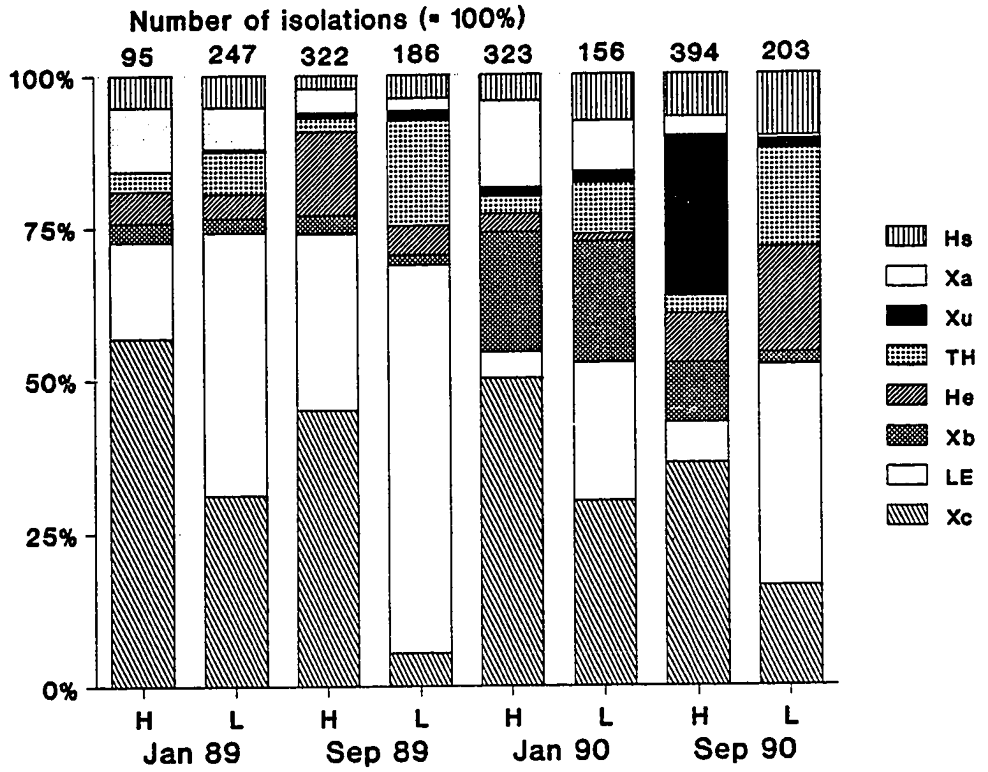


Fig. 13. Composition of the fungal communities at high (H) and low várzea (L), during the four sampling periods.

Xc	*Xylaria cubensis*	TH	*Thozetella* sp.
LE	*Letendraea* sp.	Xu	*Xylaria curta*
Xb	*Xylaria arbuscula*	Xa	*Xylaria adscendens*
He	*Hypoxylon serpens*	Hs	*Hypoxylon stygium*

3.2. *XYLARIA* SPECIES

3.2.1. Cultural studies and isozyme analysis

The purpose of the isozyme analysis in this study was to examine the degree of interspecific variation among isolates of *Xylaria* species that represented the range of morphological variation observed in pure cultures, and to determine their taxonomic relationships.

Electromorphs of all enzymes exhibited a single band in all isolates examined, which suggests a haploid organism with a single isozyme locus. There were 45 distinct electromorphs distributed among 13 species and five enzyme systems, which were found in 45 different combinations of electromorphs or phenotypes. From seven to twelve electromorphs occurred for each enzyme system. The phenotypes are listed in Table 13 by species and isolate numbers. As shown in Table 13, most isolates presented enzyme activity in the five enzyme systems used. In general two to eight phenotypes were found for each species. Species that showed the highest phenotypic diversity were isolates identified as *X. adscendens*, *X. curta*, *X. multiplex* (with five distinct phenotypes) and *X. telfairii* (with eight distinct phenotypes).

Results of the cluster analysis, following the UPGMA method, are presented in Fig. 15 showing the distance relationships among the eighty-one isolates. Four main clusters were recognized. The first cluster containing three subclusters formed by *X. microceras*, *X. arbuscula*, and *X. coccophora*. Within the second cluster, two subclusters containing *X. cubensis* and *X. castorea* respectively, were distinguished. The third cluster was represented by five subclusters constituted by *X. palmicola*, *X. multiplex*,

Table 13. Isozyme phenotypes with identity of electromorphs (**as shown in Fig. 14**) assigned to *Xylaria* species.

Phenotype	Strain #	Enzyme				
		ALD	PGI	PGM	6PG	TPI
X. adscendens						
A^1	39	6	3	8	3	12
B^3	22/31	6	7	8	3	12
C^2	7	6	7	8	1	12
D^3	41	6	7	7	3	12
E^3	34	6	2	8	3	9
X. allantoidea						
F^3	52	3	5	1	4	1
G^3	2/66	3	5	1	5	13
H^3	15	3	5	3	4	1
I^3	14/1	3	4	1	5	13
X. anisopleura						
J^2	56	6	5	10	2	11
K^3	138/137	5	5	10	5	11
X. arbuscula						
L^2	55/77/82	4	6	6	4	12
M^3	83/70/78/85/86/88	4	6	6	4	9
X. castorea						
N^2	4/47/48	4	7	3	2	2
O^2	53/65	4	7	3	5	3
X. coccophora						
P^3	58	1	6	4	3	7
Q^3	33/59/75	1	6	5	3	7

Table 13. (cont.)

Phenotype	Strain #	Enzyme				
		ALD	PGI	PGM	6PG	TPI
X. cubensis						
R[3]	3	4	5	4	3	3
S[3]	29	4	7	4	3	3
T[3]	8/20/23/63	4	9	4	3	3
X. curta						
U[2]	13/72/64/73/74/87/35/50/62	6	1	6	2	12
V[2]	6	6	1	6	2	10
W[3]	71/68	6	3	9	2	12
X[3]	49	6	3	3	2	12
Y[2]	84	6	3	4	2	12
X. ? microceras						
Z[2]	11	9	10	9	6	12
AA[2]	10	7	6	n.a.	6	12
AB[2]	40	4	3	1	6	12
X. multiplex						
AC[1]	38	6	2	7	5	12
AD[2]	76	6	4	6	5	11
AE[2]	17/21/43	6	4	11	5	n.a.
AF[2]	24/79	6	4	11	5	12
AG[2]	80	6	4	12	5	12

Table 13. (cont.)

Phenotype	Strain #	Enzyme ALD	PGI	PGM	6PG	TPI
X. obovata						
AH[2]	36	6	3	9	1	9
AI[3]	32/42/67	6	3	10	2	12
X. ? palmicola						
AJ[2]	12/5	6	7	2	1	6
X. ? telfairii						
AK[2]	18	5	3	5	4	4
AL[2]	27/60	5	4	5	7	4
AM[2]	25	5	4	6	4	4
AN[2]	26	5	4	5	4	4
AO[2]	30/133	10	5	5	4	4
AP[1]	9	5	4	10	3	4
AQ[1]	16	5	4	10	3	8
AR[1]	46	n.a.	4	10	n.a.	n.a.

[1] phenotype of ascospores isolates.

[2] phenotype of non-conidial strains.

[3] phenotype of conidial strains.

n.a.= no enzyme activity.

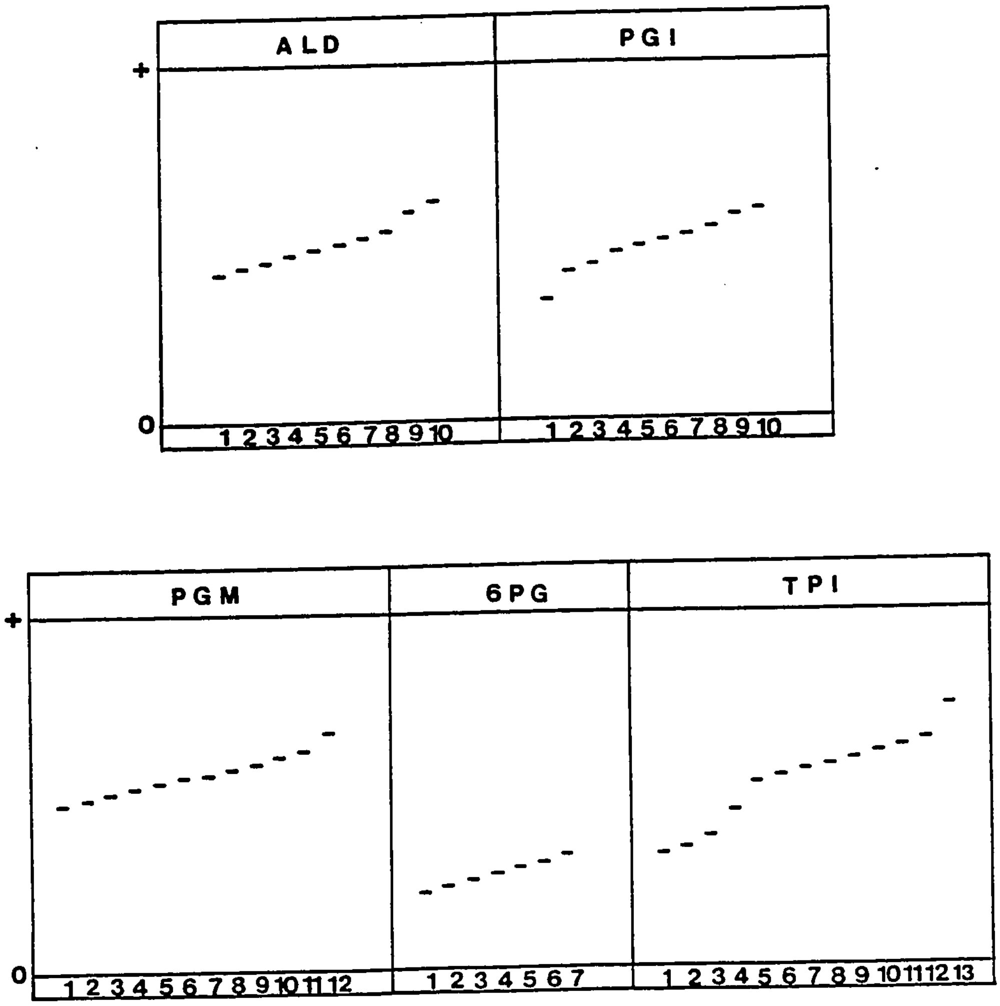


Fig. 14. Diagrammatic representation of the electromorphs of five enzyme systems found in 80 isolates of *Xylaria* species.

X. adscendens, *X. curta*, and *X. obovata*. *Xylaria anisopleura*, *X. telfairii*, and *X. allantoidea* constituted the three subclusters within the fourth and last cluster formed.

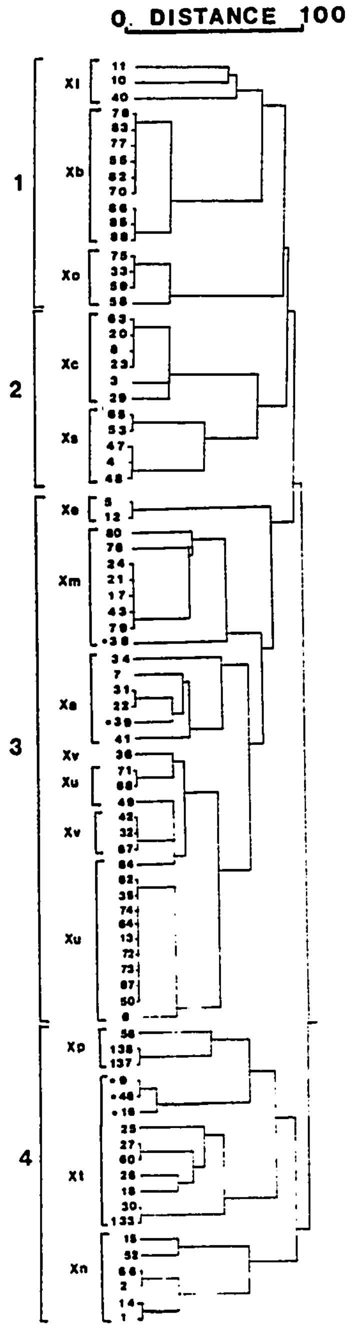


Fig. 15. Dendrogram based on isozyme phenotypes showing the similarities among *Xylaria* species isolates. Distance is normalized percent of disagreement, analyzed by the average linkage method of clustering. Xi *X. microceras*, Xa *X. adscendens*, Xb *X. arbuscula*, Xv *X. obovata*, Xo *X. coccophora*, Xu *X. curta*, Xc *X. cubensis*, Xp *X. anisopleura*, Xs *X. castorea*, Xt *X. telfairii*, Xe *X. palmicola*, Xn *X. allantoidea*, Xm *X. multiplex*.

3.3. *XYLARIA CUBENSIS*-COMPLEX

3.3.1. Isozyme and numerical analysis

The main purpose of carrying out isozyme electrophoresis with strains identified as *X. cubensis* was to determine the extent of intraspecific variation among the isolates, as well as to demonstrate the genetic diversity within species.

Fifteen phenotypes were assigned to the isolates belonging to the *X. cubensis* complex (Table 14). The greatest variation was seen in phosphoglucose isomerase patterns, which displayed four electromorphs. Conversely, all isolates showed a single, common band of 6-phosphogluconate dehydrogenase activity.

The numerical analysis was performed on cultural and biochemical data as shown by the character state matrix displayed in Appendix 5. Although two statistical techniques (cluster analysis and principal component analysis) had been performed on these data, the best graphical display for this analysis was the cluster. The dendrogram derived from the cluster analysis yielded three clusters (Fig. 17). The PCA showed that the percentage of total variance explained by the first three components was 88.2%, indicating that the model conformed to the data well. In addition, the PCA determined the characters that were important for the analysis, as follow: isozyme analysis, growth rate, pigment, and zonation.

Table 14. Isozyme phenotypes with identity of electromorphs (**as shown in Fig. 16**) assigned to *Xylaria cubensis*.

		Enzyme					
Phenotype	Ref. #	ACO	MDH	6PG	PGI	PGM	TPI
A	101/104/105/ 109/112/113/ 114/116/119/ 127/131/8	2	1	1	4	2	2
B	102	2	2	1	4	2	2
C	103	1	1	1	2	2	2
D	3	1	1	1	1	2	2
E	106	2	1	1	4	1	2
F	107/115/117/ 120/126	3	1	1	4	2	2
G	108	2	1	1	3	2	1
H	110	2	1	1	3	3	2
I	23	n.a.	1	1	4	2	2
J	118/124/129	2	1	1	3	2	2
K	121/128	2	1	1	1	2	2
L	122	2	1	1	4	3	2
M	123	1	1	1	3	2	2
N	29/125	3	1	1	3	2	2
O	130/132	1	1	1	4	2	2

n.a.= no enzyme activity

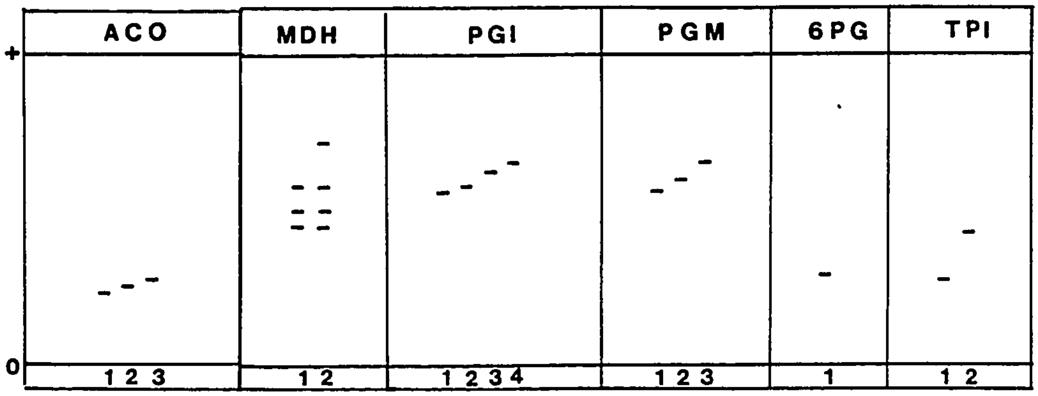


Fig. 16. Diagrammatic representation of the electromorphs of six enzyme systems found in 35 isolates of *Xylaria cubensis*.

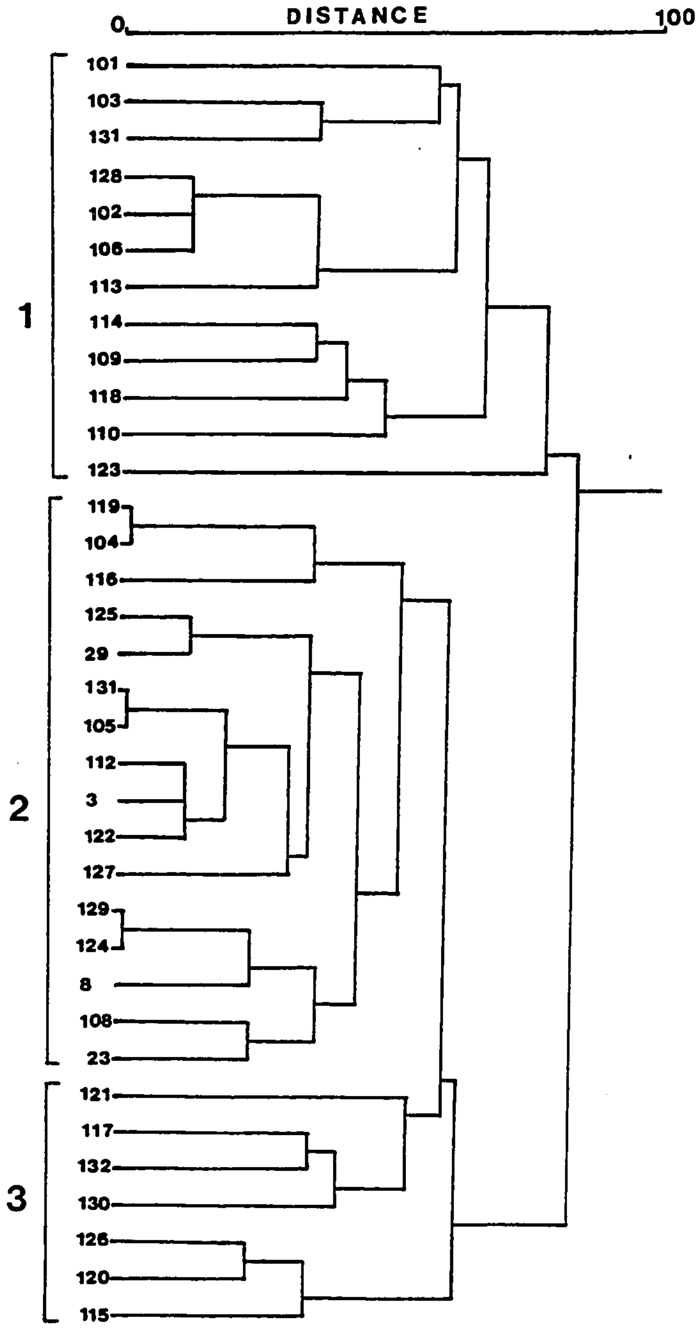


Fig. 17. Dendrogram based on cultural, and biochemical data showing the similarities among *Xylaria cubensis* isolates. Distance is normalized percent of disagreement, analyzed by the average linkage method of clustering.

3.3.2. Vegetative Compatibility Group Test

Strains of *Xylaria cubensis* used in the VCG analysis were chosen from those that occurred in the same and in different leaves of trees and seedlings that were growing in the same or nearby plots (Table 9).

In *X. cubensis* mycelial interactions resulted in either intermingling, which indicated somatic compatibility, or formation of an interaction zone, indicating somatic incompatibility. Somatic compatibility or incompatibility are reflections of genetic homogeneity or diversity, respectively. Incompatibility between strains was manifested by the formation of dark pigmentation at the line of contact of the two strains and was scored as **0**, whereas vegetatively compatible strains showed no interaction zone at contact and were scored as **1**. All self crossings were vegetatively compatible, producing a uniform mycelial mat. Strains that interacted with each other (compatible), were assigned to the same group, herein called vegetative compatibility group, or VCG. Somatic incompatibility reactions observed in strains of the *X. cubensis* complex could be assigned to the two categories reported by Sharland and Rayner (1989a; 1989b) for *H. serpens* (Pers.: Fr.) Kickx and *H. multiforme* Fr., viz. demarcation zones characterized by the formation of a "narrow line", and a "wide band", respectively.

The VCG analysis was evaluated by grouping selected isolates, according to the results from the cluster analysis (Fig. 17) performed on cultural and biochemical data presented by the same *X. cubensis* strains used in the crossings. Thus, the first cluster contained the isolates # 2, 5, 7, 10, 12, 14, 15, 19, 21, 24, 29, and 34, which made up the first subgroup for

VCG analysis (Appendix 6 a). The following vegetative compatibility groups were observed within this first subgroup VCG groups were produced:

Group 1: 2, 5, 7.
Group 2: 12, 19, 24.
Group 3: 14, 21.
Group 4: 15, 29, 34.
Group 5: 10.

Following the same procedure, the second subgroup was formed by the isolates # 1, 3, 4, 6, 8, 13, 16, 17, 22, 23, 26, 27, 30, 31, 33, 35, as shown in the Appendix 6 b. The VCG groups were as follow:

Group 6: 1, 8.
Group 7: 3, 6, 22.
Group 8: 23, 26, 33, 35.
Group 9: 13, 17.
Group 10: 16, 30.
Group 11: 31.
Group 12: 27.
Group 13: 4.

The third subgroup was formed by the isolates # 9, 11, 18, 20, 25, 28, 32, as shown in Appendix 6 c. The VCG groups were:

Group 14: 9, 18.
Group 15: 20, 32.
Group 16: 1, 28, 25.

Of the 35 isolates crossed in all possible combinations, 16 groups of vegetatively compatible isolates were found. An attempt to correlate isozyme electrophoresis with VCG techniques was made. However as shown in Table 15, most of the isolates that had the same isozyme phenotype did not always belong to the same VCG.

Table 15. Comparison among phenotypes and VCG groups found among isolates of *Xylaria cubensis*.

Phenotype	VCG isolate ref.#	VCG Group
A	4,5,6,7,13,15,23, 24,26,29,31,35	1,2,7,8,9,11
B	19	2
C	34	4
D	27	12
E	14	3
F	16,25,18,9	10,13,15
G	30	10
H	21	3
I	33	8
J	2,17	1,9
K	11,12	15,2
L	3	7
M	10	5
N	1,8	6
O	20,32	14

4. DISCUSSION

4.1. Overall colonization and endophytic communities

The presence of internal fungi in symptomless leaves of *Euterpe oleracea* was clearly demonstrated by the surface sterilization technique combined with culture work. This is the second report on the incidence of endophytes in Palmae, and the first report for *E. oleracea.* These investigations further support the assumption raised by Petrini (1986) that endophytic fungi are widespread and that all living plants most likely harbor endophytic fungi.

The low overall fungal colonization rates found in *E. oleracea* leaves is similar to that reported by Carroll and Carroll (1978) for the conifers *Abies amabilis* (20.4 %), *A. lasiocarpa* (21.1 %), *Picea breweriana* (36.3 %), *Taxus brevifolia* (24.6 %), and *Tsuga heterophylla* (39 %) in western Oregon and southwestern Washington. These authors suggested that conifers in the Pacific Northwest which typically occur only in high elevation sites might be poor hosts for blade fungi, with the result of a low overall infection rates. Rodrigues and Samuels (1990), in their preliminary investigation of endophytic fungi within leaves of the Australian palm *Licuala ramsayi* (Muell.) Domin., also found low overall colonization rates, however, no attempt was made to explain such pattern. The possible reason for the low overall frequency of colonization found in leaves of *E. oleracea* was in part due to the size of the palm leaf in relation to the number of discs sampled, i. e., a palm leaf possess a large area, thus reducing the probability of isolating dispersed fungal infection units present within the leaf.

4.1.2. Discussion of selected endophytic fungi

The two major fungal colonizers of *Euterpe oleracea* leaves were *Xylaria cubensis* and *Letendraea* sp. A pattern of dominance by one or two fungi over the entire species composition of an endophytic community recovered from a given host has been found in a wide range of vascular plants examined to date. This has been referred to as a sign of coevolution of the dominant fungi and their hosts (Carroll 1988). For instance, *Idriella licualae* K. F. Rodrigues & Samuels was the most frequent endophyte of *Licuala ramsayi* (Muell.) Domin. (Rodrigues and Samuels 1990). Similarly, *Lophodermium piceae* (Fuckel) Höhn. is dominant in *Picea abies* Karst. (Sieber 1988; Suske and Acker 1990), *Phaeosphaeria nodorum* (Müller) Hedjaroude in *Triticum aestivum* L. (Riesen and Sieber 1985), *Phyllosticta multicorniculata* Bissett et Palm in *Abies balsamea* (L.) Mill. (Petrini et al. 1989), *Apiognomonia errabunda* (Rob.) Höhn in *Fagus sylvatica* L. (Sieber and Hugentobler 1987), and *Pleuroplaconema* sp. and *Cryptosporiopsis abietina* Petrak in *Sequoia sempervirens* (D. Don ex Lamb.) Endl. (Espinosa-Garcia and Langenheim 1990).

The endophytic species isolated from leaves of *E. oleracea* comprised organisms that could be classified as "latent saprotrophs", "latent pathogens", and a "possible mutualistic symbiont". For instance, *Xylaria cubensis* and *Letendraea* sp. were recovered in large numbers and were consistently isolated over the four samplings, thus proving to be well adapted to an endophytic habit. *X. cubensis* has been reported as commonly found in decayed wood (Rogers 1984) having a cosmopolitan distribution. It has been reported to be endophytic in *Licuala ramsayi* (Rodrigues and

Samuels 1990), and in *Chamaecyparis thyoides* (L.) B. S. P. [Atlantic white cedar] (Bills and Polishook 1992), thus clearly suggesting its lack of host specificity. No species of *Letendraea* has been reported to be endophytic. In view of the high frequency in which *X. cubensis* was found within leaves of *E. oleracea,* and because of the lack of reports of disease symptoms caused by this fungus in any plant, a mutualistic relationship is speculated with the palm studied. Benefits to the host plant such as antagonism towards pathogenic fungi or decreased susceptibility to grazing insects might be suspected. *Xylaria cubensis* fulfilled four out of five requirements proposed by Carroll (1988) to recognize a possible mutualist. **1**) *X. cubensis* was shown to be widespread in *E. oleracea*, since it was isolated in great numbers from ten trees and ten seedlings sampled, in addition it was also found in another palm, *Licuala ramsayi*, suggesting a broad geographic range; **2**) although there is no report of *X. cubensis* as being seed-borne, lateral transmission presumably occurs, as shown by the VCG test; **3**) it showed the highest colonization frequency in all three types of leaf tissue examined; **4**) it has been reported to produce compounds with possible antibiotic activity (Edwards et al. 1990).

The presence of some species known to cause diseases in palms and in other tropical plants and which have been isolated as endophytes from *E. oleracea* include *Colletotrichum gloeosporioides* (Chase 1991), *Fusarium oxysporum* (Ohr 1991), *Lasiodiplodia theobromae* (Ram 1990), and *Pestalotiopsis palmarum* (Simone 1991). The occurrence of fungal pathogens in healthy plant tissues has previously been reported from a variety of plants (Sieber et al. 1988; 1991; Petrini et al. 1989; Fisher and

Petrini 1992), and confirms the assumption that a symptomless endophyte might become symptomatic when the host is under stress (Millar 1980; Andrews et al. 1985).

4.1.3. Leaf age effects

A remarkable increase in fungal colonization frequencies was consistently observed in mature leaves [leaf age 2 and 3] (Table 11). This increase could be explained by the following possibilities: 1) that the fungi were already present at an early stage in leaf development, and their mycelia gradually spread as the leaf expanded, consequently older leaves would present higher inoculum densities, and therefore they would be recovered more frequently; 2) arrival of new inoculum units. The high genetic diversity among the isolates as revealed by the isozyme analysis and by the vegetative compatibility pairings of *Xylaria cubensis* fully supports this assumption of a constant "new infection", i.e., a continuous inoculation by spores. A more detailed discussion of vegetative compatibility group systems will be presented below.

The higher number of fungi (i.e. both isolates of previously established species and of different species) recovered from older than younger leaves (Table 11) seems to be a common trend among endophytes, and has also been reported by Millar and Richards (1975), Bernstein and Carroll (1977), Petrini et al. (1979), Petrini and Müller (1979), Petrini and Carroll (1981), Oberholzer (1982), Widler and Müller (1984), Riesen and Sieber (1985), Fisher et al. (1986), Stone (1986), Bertoni and Cabral (1988), Espinosa-Garcia and Langenheim (1990). Changes in the leaf micro environment (leaf cuticle wetness, wounds, insect attack etc.) [Carroll (1979) and Foster (1977)], plant biochemical characteristics such as mineral and carbohydrate contents of leaves, and secondary compounds (Espinosa-Garcia and Langenheim 1990) have been proposed as possible physiological causes for an increase in endophyte frequency as the leaf ages.

4.1.4. Leaf tissue-type effects

The distribution of endophytes within a particular region of the plant tissue was clearly seen, and might reflect their capacity to utilize a specific substrate. In addition, the high recovery of xylariaceous fungi from the midrib regions could probably be explained by some tendency for taxonomically closely related species to share a common physical habitat.

Cases of tissue specificity in relation to vein and intervein have also been reported by other workers, e.g., Luginbühl and Müller (1980), Widler and Müller (1984), Rodrigues and Samuels (1990). However, the preference of an endophyte for a distinct location in the host tissue is not yet clear, and is one among several aspects of the complex fungus-fungus and fungus-host interactions that needs to be more rigorously inspected.

4.1.5. Tree age, and subsite effects

In general, seedlings showed higher colonization rates than trees (Figs. 11, 12, and Appendices 3, 4), revealing a tendency to harbor a larger number of endophytic isolates. This could be explained by the different proportion of sampled tissue, i.e., 60 discs from a seedling leaf represented a relatively great portion of the leaf biomass, while 60 discs from a tree leaf represented only a small fraction of a single frond.

In general, more fungi could be isolated from the high várzea than from the low várzea, and some species characterized a subsite. For example, *I. euterpes*, *Letendraea*, and *Thozetella* were characteristic endophytes of the low várzea; *X. cubensis* and the majority of the xylariaceous fungi were typically present at the high várzea. The high presence of certain species in the low várzea, where *E. oleracea* and few other palms were more frequent could be interpreted as a fair degree of host specificity presented by these endophytic species, at least at family level (Palmae), since the increase of the host (or families of the host plant) would represent an increase in substrate potential for those host specific species. In the high várzea, the floristic composition was characterized by a large diversity of species, and rarely included palms.

4.2. *XYLARIA* SPECIES

4.2.1. Cultural studies and isozyme analysis

The isozyme analysis showed considerable variation among the putative species examined. There were cases in which isolates presumed to be of one species shared no electromorphs, indicating the genetic diversity of the isolates. This phenomenon has been reported to occur in congeneric species of unrelated fungal genera, including *Atkinsonella* (Leuchtmann and Clay 1989), *Phytophtora* (Oudemans and Coffey 1991), and *Phyllosticta* (Leuchtmann et al. 1992). On the other hand, no variation was observed in isozyme patterns of isolates identified as *X. palmicola*, although it could probably be due to the small number of the fungal isolates. The isozyme banding patterns presented by isolates of *Xylaria* species confirmed the initial delimitation of most species using cultural morphology. However, some groups of isolates that were thought to represent distinct species on the basis of cultural characters were revised in the light of identical isozyme patterns.

The results of the cluster analysis (Fig. 15) suggested some relationships among the taxa investigated. For instance, the positioning of *X. curta* and *X. obovata* indicated their close affinity as previously reported by Rogers (1985) and Callan (1988), who considered these two species to belong to the *X. polymorpha* complex. Although *X. anisopleura* has also been considered as belonging to the *X. polymorpha* complex, its isozyme profile placed in a separated cluster. *X. anisopleura* was distinguished in culture by the presence of coiled hyphae in the aerial mycelia, morphology of the conidiogenous structures, stromata shape, and production of a bright orange exudate along the length of the stromata. Cultures of *X. obovata* presented colonial morphology and conidiogenous structures typical of the species as described by Callan and Rogers (1990). Isolates identified as *X. curta* were divided among two morphologically distinct groups. Isolates of one group formed stromata abundantly, and produced conidia. Isolates of the second group produced stromata scattered at the periphery of colonies, and cultures were always sterile. This variation in culture was reflected in the isozyme analysis, in which isolates presented different enzymatic activity as shown by the five phenotypes produced. Conidial strains of *X. curta* were represented by three phenotypes (W, X, Y), whereas non-conidial strains presented two phenotypes (U, V).

Xylaria arbuscula and *X. coccophora* have been considered as related to the *X. multiplex* complex by Callan (1988), and the inclusion of *X. microceras* in the same complex was suggested by Joly (1968). According to the results of the cluster analysis, the three species, *X. arbuscula*, *X. coccophora*, and *X. microceras*, appeared to form a closely related group of species based on their isozyme profiles. However, endophytic strains as well as ascospores isolates identified as *X. multiplex* did not group together with the three species mentioned above. Cultures of *X. arbuscula* were basically characterized by the production of black stromata with white apices that form in concentric rings, morphology of the conidiogenous structures, which were produced either on the cylindrical stromata or on pulvinate structures formed on the colony surface. Cultures of *X. coccophora* were characterized by the production of a canary yellow pigmentation on the surface of the colony and stromata. This, along with the production of thin, wiry stromata and morphology of conidiogenous structures, is in accordance with the descriptions of *X. coccophora* given by Callan and Rogers (1990). *X. microceras* produced neither stromata nor conidiogenous structures in culture. The identification of this species was only possible by comparison with isolates growing at WSU that were derived from cultured teleomorphs collected in Venezuela.

Although *X. cubensis* has never been considered to be related to *X. castorea*, the two species were clustered close to each other. Endophytic strains of both species shared the unique feature of salmon colored mycelium in cultures. However, the absence of conidiogenous structures, and the predominantly cylindrical stromata that were broad and villose at the base, and gradually tapered towards the apex clearly distinguished *X. castorea* from *X. cubensis*.

Cultures of *X. telfairii* were characterized by the following features: colony color, presence of coiled hyphae in the aerial mycelium, and stromata fan-like shape. All endophytic strains were sterile, also in accordance with previous reports on this species in culture given by Callan and Rogers (1990). Although cultures of the three ascospores isolates of *X. telfairii* derived from different hosts were morphologically similar to the endophytic isolates, the isozyme profiles of cultured teleomorphs were not comparable to the endophytic strains. Isolates of *X. telfairii* derived from ascospores clustered close to endophytic isolates of *X. anisopleura.*

X. adscendens, a species closely allied to *X. hypoxylon* (L. : Fr.) Grev. (Rogers 1984), was frequently isolated as an endophyte. The colony appearance of *X. adscendens* isolates presented a typical canary yellow pigmentation which soon turned olive-black. Endophytic strains identified as *X. adscendens* were characterized by four phenotypes, while cultured teleomorphs presented different enzymatic activity.

The results indicate that isozyme electrophoresis, when used in combination with cultural characters, is a useful supplementary tool to detect variations at intraspecific level among endophytic species of *Xylaria.* However, as no similarities were detected between isozyme activities of cultured teleomorphs and endophytic isolates, this technique proved to be inadequate when used in combination with isozyme data from ascospore isolates of identified species derived from different hosts.

4.3. *XYLARIA CUBENSIS*-COMPLEX

4.3.1. Isozyme and numerical analysis

The results of the numerical analysis, that included cultural and isozyme studies, showed that isozymes, and three features of the colony morphology (growth rate, diffusing pigment and zonation) were important in defining different morphotypes. However, these attributes did not favor a separation into distinct taxa, they rather indicated that a more precise taxonomical determination should include not simply the conidial but also data from the ascigerous state. Investigation of such a "species complex" from the taxonomic and evolutionary point of view is needed because of the frequency with which members of the Xylariaceae are encountered as endophytes.

4.3.2. Vegetative Compatibility Group test

The importance of performing VCG tests for establishing genetic diversity through mycelial interactions has been demonstrated by Chapela et al. (1988), Chapela and Boddy (1988a; 1988b), Elias et al. (1991), and Rayner (1991a; 1991b).

Mycelial interactions have been reported from works on endophytic fungi of *Eucalyptus viminalis* Labill. (Faifer and Bertoni 1988), *Fagus sylvatica* L. (Chapela and Boddy 1988a; 1988b; Chapela et al. 1988), and of *Pseudotsuga menziesii* (Mirb.) Franco (Sherwood et al. 1986) in order to assess genetic diversity. Mycelial interactions in xylariaceous fungi have been investigated with *Daldinia concentrica* Ces. & DeNot. (Sharland and Rayner 1986), *Rosellinia desmazieresii* (Berk. & Br.) Sacc. (Sharland et al. 1988), and *Hypoxylon* species (Chapela and Boddy 1988a; 1988b; Sharland and Rayner 1989a; 1989b).

In general among the *Xylaria* endophytes of *Euterpe oleracea* there were at least 2 different VCGs in the same leaf and different leaves of the same tree or seedling, indicating that an individual tree is infected by more than one strain, thus there were different populations of *X. cubensis* in a single tree. This is demonstrated by the high degree of somatic incompatibility observed, which reflects a great genetic diversity. Results from the VCG analysis suggest that the pattern of colonization presented by *X. cubensis* in leaves of *E. oleracea* is characterized by numerous genetically distinct inoculum units sparsely allocated. This spatial pattern of endophytic distribution has been referred to as "tight compartments" and it seems to be common among some endophytic species such as *Apiognomonia errabunda* in beech leaves (Petrini 1991) and *Rhabdocline parkeri* in Douglas fir needles (Stone 1987).

5. TAXONOMIC TREATMENT

Hawksworth et al. (1983), and Rossman et al. (1987) were consulted as the primary sources for taxonomic literature. The classification of the Classes Loculoascomycetes and Hymenoascomycetes (Ascomycotina) followed Barr (1987; 1990, respectively).

Cultural characteristics of xylariaceous fungi were studied on colonies growing on 9 cm Petri dishes containing Oatmeal agar (OA, Appendix 1), except for *Virgariella*. The plates were inoculated with approximately 4 x 4 mm blocks of mycelium taken from the edge of actively growing colonies on CMD. Because cultures were sterile or presented only the anamorph they could be identified only through comparison with colonies known to have originated from ascospores of identified species. Although perithecial production in *Hypoxylon* has been reported to occur in colonies growing on Scratch Malt agar (Kenerley and Rogers 1976), my isolates failed to produce any ascomata after several attempts and after long incubation, thus *Hypoxylon* species were further studied on OA.

The Deuteromycotina was divided into the two classes, Coelomycetes and Hyphomycetes. The classification of Coelomycetes followed Sutton (1980), in which class and subclass divisions are defined according to the origin of conidia (blastic or thallic) and the conidium wall (holoblastic x enteroblastic; holothallic vs. enterothallic).

Isolates identified as *Phomopsis* were studied on Czapek-Dox (CZ, Difco) to allow production of diagnostic characters, including pigmentation, as previously reported by Rodrigues and Samuels (1990).

Gross colony morphology of *Fusarium* species was studied on Potato Dextrose agar (PDA, Difco); conidial characters were taken from SNA. Identification of *Fusarium* species followed the scheme of Gerlach and Nirenberg (1982).

Numbers appearing inside parenthesis in the section "**ISOLATES EXAMINED**," represent culture tube numbers that are deposited in the culture collection at NY. Representative isolates for each species that were sent to ATCC are preceded by the letters KFR.

5.1. ASCOMYCOTINA

LOCULOASCOMYCETES

PLEOSPORALES:

TUBEUFIACEAE

Letendraea **sp.** (Plate I, Figs. 1-2)

CMD: Colony 1 cm diam within 5 days, with little aerial mycelium; white to pale, then pale orange (5A3) from the point of inoculum outwards, azonate; margin white, even. **PDA**: colony 1.5 cm diam within 5 days; cottony, zonate; carrot red (6B7) darkening to brownish orange (6C8); margin, white, entire; reverse brownish orange (6C8). Ascomata produced after 10 days of incubation at center of colony; superficial, scattered; colorless first, then yellowish; globose to subglobose, smooth, glabrous, 222.5-292.5 x 267-292.5 µm. Cellular pseudoparaphyses few, to 90 µm long x 3.6 µm wide, unbranched, septate, deliquescing in age. Asci few, broadly clavate, nearly sessile, (63-)62.6-78.4(-81) µm long x (19.8-)22.3-38.6(-38.7) µm wide, bitunicate, apex with a "nasse apicale." Ascospores oblong, (14-)19.2-26.4(-27.5) x (7.2-)8.2-13.3(-14.7) µm, two-celled, septum submedian, each cell with one to three globules, hyaline, smooth, partially biseriate. Anamorph not observed.

Total number of isolates: 483.

Representative isolates at ATCC: 89-30, 91-527, 91-564, 91-580, 91-582.

COMMENTS: This is the second most commonly isolated fungus. It occurred chiefly in seedlings, and was present at both subsites, but with higher frequency in the low várzea. It occurred in the midribs and intervein tissues, but more frequently in the later, and was equally common in all of the three leaf ages sampled.

This is likely to be an undescribed species. The two species of *Letendraea* accepted to date (Barr 1980; Rossman 1987) have smaller ascospores and narrower asci.

ISOLATES EXAMINED: **Jan 89**: 27i1b; 27i2b (89-32); 27i2c; 27i2d; 30i1c; 30i1d; 30i1e; 30i2d; 57i4b; 60ia; 60v1c (89-30); 60v3a; 63i3a1; 63i3b; 63i3c1; 63i3e; 63i4c1; 63i4d; 63i4e2; 66i1a; 66i1b; 66i2c; 66i2d; 66v1c; 66v1d; 67i2a; 69i1a; 69i1d; 69i2c; 69v3c; 71i1a; 71i1b; 71i1c; 71id1; 71i2a; 71i2b; 71i2d; 71v1b; 71v1c; 71v1e; 71v2b; 73i3a1; 73i3b; 73i3c; 73i3d; 73i3e; 73v1c; 73v1d; 75i1a2; 75i1b; 75i1c; 75i1e; 75i2a1; 75i2b; 75i2c; 75i2d; 75i2e; 75v1d; 75v1e; 84v1d2; 86i1b; 86i1c1; 86i2a1; 86i2b1; 86i2b2; 86i2c1; 86i2c2; 86i2d1; 86i2d2; 86i2e1; 86i3a2; 86i3b; 86i3d1; 86i3e; 88i2c; 88i3a1; 88v2c; 88v2e; 90i1c; 90i1e; 90i2b; 90i2c; 90i2d; 90i2e; 90i3a; 96i2a; 96i2b; 96i2c; 98i1a; 98i1b; 98i1c2; 98i2c; 98v2a; 100i2b; 100i2c; 103i1a1; 103i1a2 (89-138); 103i2a; 103i2b; 103i2c; 103i2d; 131i2c; 131v1a; 133v2a; 133v2b; 156v1a; 169i1a (89-43); 169i1b; 169i1c; 169i2a; 169i2b; 169i2c; 171i1a; 176i1a1; 184i1b2; 184i1c; 184i2c; 184v2d; 200i1c2; 200i1d; 223i1a.

Sep 89: 7v2a; 8v2a; 9v1a; 14v2a; 29i2b; 37v1c; 41v1c; 56v1a; 66i2a; 67v1e; 68i2a; 69i2a; 69v1a; 91i1d; 91i2b; 91i2c; 91i2d; 91i3c; 91i3d; 91i4b; 91i4c; 91i5b; 91i5c; 91i5d; 91i6a; 91i6b; 91i6c; 91v3c; 92i1c1; 92i1d1; 92i1e; 92i2a2; 92i2c; 92i2d; 92i2e; 92i4d; 92i5b; 92i5c; 92i6c; 92v4b; 92v4c; 92i6e; 94i2b; 94i2c; 94i4a; 94i4b; 94i4c; 94i6a; 95i1a; 95i1b; 95i1c; 95i2b;

95i2c; 95i3a; 95i3b; 95i3c; 95i4b; 95i6b; 95v5d; 97i2d; 97i3b; 97i4a; 97i4d; 97i5b; 98i1a; 98i1b; 98i1c; 98i1d; 98i2a; 98i2b; 98i3a; 98i3b; 98i3c; 100i1b; 100i5c; 101i5b; 102i6a; 103i1a; 103i1b; 103i2a; 103i2b; 103i2c; 103i3b; 103i3c; 103i5a; 103i5b; 103i5c; 103i5d; 103i6b; 103i6c; 103v3b; 103v3d (90-53); 103v5d; 104i1a; 104i1b; 104v3b; 105v3b; 106i1a; 106i1b; 106i1c; 106i2a; 106i2b; 106i2b; 106i3a; 106i3b; 106i3c; 106i3d; 106v3a; 107i1a; 107i1b; 107i2a; 107i2b; 107i2c; 107i2e; 107i3a; 107i3b; 107v1b; 107v2c; 107v3a; 107v3b2; 108i1a; 108i2a; 108i2b; 108v3e; 109i1b (91-580); 109i1c; 109i1d; 109i1e; 109i2a; 109i2b; 109i2c; 109i2d; 109i3a; 109i3b; 109i3c; 109i3d1; 112i1c; 112i3b; 112i3c; 112i3d2; 112v1b; 112v3d; 113i1c; 113v1e; 114i3b; 114i3c; 115i1a; 115i1b; 115i1c; 115i3b; 115i3c; 115i3d; 115v2d; 118i1a; 118i1b; 118i2b; 118i2c; 118v1b; 118v3b; 118v3c; 119i1a; 119i2b; 119i2d; 119i3b; 120i1c; 120i1d; 120i2a; 120i2b; 120i2c; 120i3a; 120i3b; 120i3c; 121i1a1; 121i1b1; 121i1c; 123i1b; 123i1c; 123i2a; 123i2b; 123i2c; 123i3a; 123i3b; 123i3c; 123v2b; 123v3d; 123v3e; 124i1a; 124i1b; 124i1c; 124i3c2; 124v2e; 125i1a; 125i1b; 125i2b; 125i2c; 125v1c; 125v1d; 125v2a; 126i2a (91-578); 128i1d; 128i2e; 128i3d; 130i2a; 130i2b; 130i2c; 131i3a; 131i3b; 131v1e.

Jan 90: 298i1a; 298i1b; 298i1c; 309i2a; 311v3d; 312i1a; 312i2b; 313i1a; 317i1a; 317i1b; 317i1c; 318i2a; 318i3b; 319i1b; 319i1c; 319i3a; 319i3b; 320i1b; 320i1c; 320i2a; 324i2a; 324i2b; 332i2a; 332i2b; 332i2e; 332i6a; 335i2a; 335i2b; 335i2c; 335i5c; 335i5d; 336i4b; 343i3a; 343i3b; 343i3c; 343i4a; 343i4b; 343i4c; 343v6a; 343v6b; 344i1a; 344i1b; 344i1c; 344i1d; 344i2a; 344i6a; 344i6b; 346i3a; 346i3b.

Sep 90: 366ia; 369ia; 394ia; 394ib; 396ia; 396ib; 406ia; 407ia; 407ib; 414i3d; 414i4a (91-528); 414i4b; 414i4c1; 414i4c2; 414i6a; 414i6b; 414i6c; 414i6d2; 414v6c2; 415i4a; 415i4b; 415i4c; 415i5a; 415i5b; 415i5c; 415i6a; 415i6b; 415i6c; 415i6d; 415v4c; 415v4d (91-526); 415v6c; 415v6d; 417i4c; 417i4d; 417i4e; 417i5d; 417i5e; 418i2c; 418i2d; 420i4a; 420i4b; 420i4c; 420i4d; 420i5a; 420i5b; 420i5c; 420i6c; 420i6d; 420i6e; 421i1b; 421i1c; 421i3a; 421i3b; 421i3c; 421i4a; 421i4b; 421v6a; 421v6b; 421v6c; 424id; 424i4c; 424i5d; 427i3d (91-564); 427i3e; 428i1a (91-527); 428i1d; 428i2b; 428i2c; 428i3a; 429i1c; 430i1a; 430i1b; 432i3b; 432i3c; 432i3d; 432i6a; 435i4d; 435i5b; 435i5c; 436i4a; 436i4b; 436i5a; 436i5b; 438i5a; 438i5b; 439i4a; 439i4b; 442i6c; 442v6d; 454v1c; 465i1b; 465i2a; 474i1a; 474i1b; 479i1e; 480i2b; 483i2a; 486i1a.

PYRENOMYCETES

HYPOCREALES:

HYPOCREACEAE

Calonectria sp. (Plate II, Figs. 1-3)

Anamorph: *Cylindrocladium* (Plate III, Figs. 1-2)

OA: Colony 2 cm diam in 5 days; cottony, azonate; aerial mycelium white to orange white (5A2). Reverse non pigmented. Ascomata produced after 10 days of incubation all over the colony; superficial, orange red (8A8), turning red in KOH, and turning yellow in cotton blue, subglobose, with warty surface, 356-445 x 445-534 µm. Paraphyses not seen. Asci few, clavate with a short stalk, (72-)90-112.5 µm long x 20.7- 21.6 µm wide, unitunicate, 4-8-spored. Ascospores fusiform with rounded ends, (35.1-)46.8-54(-63) x (4.5-)5.4-6.3 µm, 1-3 septate, contents granulous, hyaline, smooth, multiseriate. Conidiophores arising from aerial mycelium, 81-135 long x 6.3-9 µm wide, hyaline, smooth, penicillately branching. Conidiogenous cells phialidic, solitary, terminal, 9-22.5 x 3.6-4.5 µm. Conidia cylindric, straight, 40.5-54-67.5 x 4.5-5.4 µm, 2-4-celled, hyaline, smooth. Chlamydospores in chains, subglobose, pale brown.

Total number of isolates: 1.

COMMENTS: This endophyte was isolated only once, from a leaf of a seedling, located at high várzea.

Several *Calonectria* species, all of which have *Cylindrocladium* anamorphs, have been reported as causal agents of leaf spots on palms (Aragaki and Uchida 1991). The current results suggest that these pathogens might have an endophytic phase sometime in their life-span.

ISOLATE EXAMINED: 442v2a (91-517).

XYLARIALES:

AMPHISPHAERIACEAE

Phomatospora sp. (Plate IV, Figs. 1-2)

CMD: Colony 1.5 cm diam in 5 days; opaque, aerial mycelium scant, faintly zonate; pale first, then honey yellow (5D6); margin colorless, even to narrowly lobed. Perithecia semi-immersed to superficial, scattered; dark brown, subglobose, papillate, glabrous, (226.9-)445-534 x (230-)311.5-489.5 µm. Paraphyses filiform, abundant, to 125 µm long x 4 µm wide, unbranched, septate, hyaline. Asci few, cylindrical, stipitate, (73.8-)82.5-93.5 µm long x 6.3-7.2 µm wide; unitunicate, apical ring J^+ Melzers, 8-spored. Ascospores ellipsoidal, (7-)12.6-14.4 x 2.7-3.6 µm, one-celled first, then 2-celled with age, septum median, eventually biguttulate, hyaline, smooth, biseriate. Anamorph not observed.

Total number of isolates: 23.

Representative isolates at ATCC: 90-88, 91-348, 91-497, 91-554.

COMMENTS: Although occurring in both trees and seedlings, this species was more frequently isolated from trees. It was equally present in both sites. It occurred in both newly expanded and mature leaves, and was isolated from midrib and intervein tissues.

ISOLATES EXAMINED: **Jan 89**: 8vd (89-18); 37v2d; 44v3a; 60i2a; 66v1e (90-120).

Sep 89: 41v1a (90-88); 56v2c; 60v1e (91-22); 68v1b; 76v1b (91-69); 76v2c (91-20).

Jan 90: 263v2b (91-487); 270v1b (91-488); 291v6a2 (91-348).

Sep 90: 362va (91-496); 396va (91-563); 420v5b; 427v3e (91-552); 442v6a1 (91-554); 444i2b (91-509); 444i5c; 454i1c (91-497); 479ia.

HYPONECTRIACEAE

Leiosphaerella cocoes (Petch) Samuels & Rossman, Mycotaxon 28: 465. 1987. (Plate V, Figs. 1-2)

OA: Colony reaching 1 cm diam in 5 days, felty to appressed with radial furrows; at first white, then birch grey (5C2) at center and whitish towards edge, margin colorless, minutely lobed. Colony not reaching full diameter of Petri dish. Reverse pompeian yellow (5C6) to oak brown (5D6), radially spliting agar. Ascomata produced around point of inoculum, semi-immersed, scattered; blackish, globose to subglobose, rostrate. Paraphyses filamentous, to 2.7 µm wide, deliquescing with age, septate, hyaline. Asci few, cylindrical, 130.5-157.5 µm long x 8-10 µm wide, unitunicate, apical ring J+ Melzers, 8-spored. Ascospores fusiform, (50.4-)63.8-88.3(-85) x 2.7-3.6 µm, 2-celled, septum median, minutely guttulate, hyaline, smooth, multiseriate. Anamorph not observed.

Total number of isolates: 8.

Representative isolates at ATCC: 89-160, 91-590.

COMMENTS: This endophyte was isolated more frequently from trees in the low várzea. All isolates were recovered from the midribs of mature leaves, with the exeception of only one isolate which came from the intervein region.

The isolates found in this study agree with the description of the species given by Samuels and Rossman (1987).

RECORDED AS ENDOPHYTE: none.

OTHER SUBSTRATA: on *Cocos nucifera* L. (fronds and fruits).

DISTRIBUTION: tropical.

ISOLATES EXAMINED: **Jan 89**: 84v1b (89-149); 100v1a (89-42); 133i1a (89-114; 89-154); 135v1b.

Sep 89: 9v3c; 10v1b (90-106); 37v2d..

Jan 90: none.

Sep 90: 366ia (91-590).

Oxydothis poliothea Syd., Ann. Mycol. 28: 91. 1930.

(Plate VI, Figs. 1-2)

CMD: Colony 1.5 cm diam in 5 days; opaque, azonate; aerial mycelium scant, olive brown (4E5); margin even, colorless. Ascomata formed around edge as well at center; superficial, gregarious; black, globose to subglobose, papillate, glabrous. Paraphyses filamentous, 1.8-3.6 µm, deliquescing with age, septate, hyaline. Asci unitunicate, cylindrical, > 90 µ m long x 10.8-11.7 µm, apical ring amyloid, 8-spored. Ascospores multiseriate, fusiform, 85.5-103.5 x 5.4-6.3 µm, 1-septate, septum median, hyaline. Anamorph not produced.

Total number of isolates: 3.

Representative isolates at ATCC: 91-572.

COMMENTS: This endophyte was isolated from seedlings located at low várzea. It was recovered from the midribs of newly expanded leaves.

RECORDED AS ENDOPHYTE: none.

OTHER SUBSTRATA: on palms: *Maximiliana*, *Orbygnia*, *Scheelea*.

DISTRIBUTION: tropical.

ISOLATES EXAMINED: **Jan 89**: 41v2d.

Sep 89: none.

Jan 90: none.

Sep 90: 415v1a2 (91-571); 418v2b (91-572).

Physalospora sp. (Plate VII, Figs. 1-2)

CMD: Colony 1.5 cm diam in 5 days; translucent, aerial mycelium scant, white; azonate, margin even. Perithecia semi-immersed, scattered, dark brown, subglobose, papillate, smooth, glabrous, about 0.2 mm diam. Paraphyses filiform, to 63 µm long x 2.7 µm wide, unbranched, septate. Asci few, clavate-cylindrical, nearly sessile, 54.6-61.6 µm x 16.1-21.6 µm, unitunicate, apical ring refractive, 8-spored. Ascospores ellipsoidal, (13.3-)14.4-17.2(-20) x (4.9-)5.3-7(-8.1) µm, one-celled, with granular contents, hyaline, smooth, partially biseriate. Anamorph not produced.

Total number of isolates: 3.

Representative isolate at ATCC: 89-135.

COMMENTS: This endophyte was isolated from seedlings at low várzea. It occurred in both tissues (midribs and intervein), and in both leaf age classes (mature and newly expanded).

ISOLATES EXAMINED: **Jan 89**: 90i1a (89-17), 103v1a (89-135).

Sep 89: none. **Jan 90**: none.

Sep 90: 418i1c.

XYLARIACEAE

Because members of the Xylariaceae are often distinguished by anamorphs that may be accomodated in the form-genera *Geniculosporium* Chester & Greenhalgh, *Nodulisporium* Preuss., and *Virgariella* Hughes, I have included these deuteromycete genera in this part, and not among fungi imperfecti.

Cultures were easily recognized as being xylariaceous because of typical features that characterize the family such as colony morphology (growth rate, color, texture, stromatic hyphae in the aerial mycelium, diffusing pigment), anamorphic structures (formation of anamorph-bearing stromata), and conidial ontogeny. However, because telemorphs rarely formed in culture, an *Anthostomella* species being the only exception, it was difficult to distinguish from among the genera of the family. Anamorphs were frequently formed, but are of limited use in generic taxonomy in the family as it is now known. For example, *Nodulisporium* anamorphs are formed by species of *Daldinia*, *Hypoxylon*, *Rosellinia*, and *Xylaria*.

Sterile colonies that lack stromal production but that formed thick walled, pigmented stromatic hyphae in the aerial mycelium (viz, coiled, reticulate, or long filaments with short protuberances), combined with other cultural characters that are typical of the family as outlined above, were treated as Xylariaceae. While most isolates could be referred to a genus, several isolates could not and these were assigned to one of five groups designated as Xylariaceae I, II, III, IV, V.

Accurate identification can be accomplished only when colonies are compared with cultures derived from ascospores from identified teleomorphs

(Petrini and Petrini 1985; Callan and Rogers 1990). An analysis that combines morphology and cultural characters with biochemical methods such as the production of secondary metabolites (Whalley and Edwards 1987; Edwards et al. 1990), DNA sequences (Rogers 1990), and isozyme profiles (O.Petrini, pers. comm.) can lead to precise identification. However, the number of species of xylariaceous fungi that have been cultured from their ascospores is very low, thus limiting the ability to precisely identify sterile or anamorphic endophyte cultures.

Anthostomella sp. (Plate VIII, Figs. 1-2)

Colony 1.5 cm diam in 5 days; felty to floccose, shallowly furrowed in a radial pattern, azonate to faintly zonate towards margin; white, occasionally turning cream (4A3) with age; margin white, narrowly lobed; colorless exudate on aerial mycelium; stromatic hyphae not seen; reverse non pigmented to honey yellow (5D6). Colony not reaching full diam. of Petri dish. Ascomata produced at center of colony, immersed, scattered, dark brown, subglobose, 195-374 x 182-352 µm. Paraphyses not observed. Asci few, cylindrical with a short stipe, (51.8-)56-65(-75) µm long x 4.2-5.6 µm wide, unitunicate, apical ring J^+ Melzers, 8-spored. Ascospores ellipsoidal, with longitudinal germ slit, (10.8-)13.5-14.9(-17) x 4.5-5.4 µm, 2-celled, small cell hyaline and appendiculate, 1.8 x 2.7 µm, bigger cell olivaceous brown, (9-)11.7-13.1 x 4.5-5.4 µm, smooth, uniseriate. Anamorph not produced.

Total number of isolates: 16.

Representative isolates at ATCC: 89-31, 89-126, 90-152.

COMMENTS: This endophyte was isolated from both trees and seedlings at high várzea. It occurred in both leaf ages (mature and newly expanded), mostly in the intervein tissue.

A taxonomic treatment of this genus was provided by Francis (1975). Four species were described from palms, however, they all have one-celled ascospores.

ISOLATES EXAMINED: **Jan 89**: 184i1b1 (89-126); 186i2b; 190i2a1; 190i2b (89-155); 192i2a; 209i2a1; 221i1d; 221i2a (89-31); 221i2b; 223i1c.

Sep 89: 97i2c (90-132); 97i4e; 104v3a (90-152); 104v3c.

Jan 90: 304i2d (91-168); 331i5c (91-169).

Sep 90: none.

Daldinia eschscholzii (Ehrenb.) Rehm, Ann. Mycol. 2: 175. 1904.
Anamorph: *Nodulisporium* (Plate IX, Figs. 1-3)

Colony 5 cm diam. in 7 days; floccose, faintly zonate at periphery; white first, then darkening to bronze (5E5); margin white, entire; stromatic hyphae reticulated (Fig. 3); reverse dark blonde (5D4); pale brown pigment diffusing. Stromata not produced. Conidiophores produced all over colony surface, to 90 µm long x 3.6 µm wide, subhyaline to pale brown, smooth, irregularly branching. Condiogenous cells solitary or whorled, terminal and intercalary, cylindrical, 9-19.8 x 2.7-3.6 µm. Conidia obovoid to ellipsoidal, (5.4-)6.3-7.2 x 2.7-3.6-4.5 µm, one-celled, with flat basal abscission scar, hyaline to pale brown, smooth.

Total number of isolates: 2.

Representative isolate at ATCC: 90-95.

COMMENTS: This endophyte was isolated both from a tree and a seedling located at both subsites (high and low várzea). It occurred in the intervein region of newly expanded leaves.

This description agrees well with the descriptions of *D. eschscholzii* given by Petrini and Müller (1986), although conidia described by these authors are larger (5-10 x 4-6µm) than those found here.

RECORDED AS ENDOPHYTE:none.

OTHER SUBSTRATA: on wood of *Celtis*, *Citrus*, *Coccos*, *Dalbergia*, *Dipterocarpus*, *Eucalyptus*, *Fraxinus*, *Hevea*, *Papaya*, *Passerina*, *Quercus*, *Tamarindus*.

DISTRIBUTION: tropical and subtropical.

ISOLATES EXAMINED: **Jan 89**: none.

Sep 89: 57i1a (90-95); 115i2a.

Jan 90: none.

Sep 90: none.

Hypoxylon quisquiliarum (Mont.) Mont., Ann. Sci. Nat. Bot., Ser. 4, 3: 117. 1855.

Anamorph: *Nodulisporium*-like (Plate X, Figs. 1-3)

Colony 2 cm diam in 5 days; cottony with barraging ring from the center; first white to canary yellow (2B7) then overlain with brown layer at center; margin white, entire; yellowish to yellowish brown exudate in aerial mycelium; reverse greyish orange (5B4-5B5). Synnemata formed within 1 to 3 weeks from the center outwards, to 2 mm tall, stipe off-white, capitulum khaki (4D5) to brown (5E5). Conidiophores forming a palisade at the apex of the synnema, 90-126 µm long x 4.5-5.4 µm wide, pale brown, smooth, dichotomously branching near the apex. Conidiogenous cells terminal, cylindrical, 13.5-27 x 5.4-6.3 µm, pale brown, smooth, with conical to denticulate secession scars. Conidia obovoid to pyriform, (9-)8.8-10.7(-13.5) x (3.6-)4.5-5.9(-6.3) µm, 0-septate, base flat, a few constricted and forming a "beak" at base, hyaline, smooth. Chlamydospores subhyaline.

Total number of isolates: 9.

Representative isolates at ATCC: 90-200, 91-73, 91-521.

COMMENTS: This endophyte was most commonly isolated from seedlings, at both sites. It occurred equally in both tissues (midribs and interveins) of mature leaves.

Hypoxylon quisquiliarum is a tropical species commonly found on palms in the Amazonian forests. In colony characters and morphological details of synnemata and conidia, this endophyte closely resembles the only known ascospore isolate of *H. quisquiliarum* (NY: Samuels 1888, GJS 84-88, unpublished).

RECORDED AS ENDOPHYTE: none.

OTHER SUBSTRATA: on angiospermous bark and wood (Rick 1931; Dennis 1963; Miller 1961); *Cocos* (Viégas 1961).

DISTRIBUTION: tropical.

ISOLATES EXAMINED: **Jan 89**: 98i2b.

Sep 89: 67i2d (91-73); 78i1a; 94v6d; 103v2a; 113v2d.

Jan 90: 313v2a (90-200).

Sep 90: 432i2a (91-472); 441i5a.

Hypoxylon serpens (Pers.: Fr.) Kickx, Fl. Crypt. Louv., p.115. 1835.
Anamorph: *Geniculosporium* (Plate XI, Figs. 1-3)

Colony 3 cm diam in 7 days; finely cottony to velvety, widely zonate; at first white to orange white (6A2), then darkening to ash grey (1B2), finally to cement (4D2) in the concentric zones; margin white, entire to scalloped; a colorless exudate forming in the aerial mycelium; stromatic hyphae dark brown (Fig. 3); reverse greyish orange (6B5); no pigment diffusing. Stromata lacking. Conidiophores arising on surface of colony, in concentric zones, > 90 μm long x 2.7-3.6 μm wide, subhyaline, smooth, dichotomously branching. Conidiogenous cells terminal and intercalary along the length of conidiophores, cylindrical, geniculate, hyaline, smooth, with circular secession scars. Conidia obovate to elliptical, (2.8-)3.5-5(-5.6) x (2.1-)2.3-3(-3.5) μm, one-celled, with a frilled basal abscission scar, few with truncate base, hyaline, smooth.

Total number of isolates: 147.

Representative isolates at ATCC: 90-127, 91-307, 91-314, 91-316.

COMMENTS: This endophyte was isolated both from trees and seedlings in both subsites. It was present in all three leaf age classes, and in both tissues, however, it was more frequently recovered from the midrib region.

This description agrees with those given by Petrini and Müller (1986), and Petrini and Rogers (1986) for *H. serpens*.

RECORDED AS ENDOPHYTE: in *Triticum aestivum* L. (Riesen and Sieber 1985), *Pinus nigra* Arnold, *Mahonia nervosa* (Petrini and Rogers 1986), *Licuala ramsayi* (Muell.) Domin. (Rodrigues and Samuels 1990), *Alnus rubra* Bong. (Sieber et al. 1991), *Chamaecyparis thyoides* (L.) B. S. P. (Bills and Polishook 1992).

OTHER SUBSTRATA: *Corylus*, *Elaeis*, *Fagus*, *Fraxinus*, *Hibiscus*, *Quercus*, *Ulmus*, *Salix*.

DISTRIBUTION: cosmopolitan.

ISOLATES EXAMINED: **Jan 89**: 30v1a (89-137); 30v1b; 39i1a2; 49i3a; 60v3d; 63v2e; 84i1c; 84i1d; 88v3c; 100v2e; 107v1b; 169v1b; 170ib (89-130); 203v2a; 216v2c; 218i1b; 223i1b.

Sep 89: 46v1a; 48v2a; 55v1b (90-36; 90-62)); 55v1d (90-58); 55v2a; 56i2a; 56v2a; 56v2b; 58i2b; 59i1a; 60i1b; 60v1a; 61v1a; 74v2a; 82v2a; 91v4c (90-119); 91v4d (90-116); 92i3c; 92v5a (90-147); 92v5b1; 94v1a; 94v2a; 94v4a; 94v4b; 95v1a; 95v3a (90-124); 97i1a (91-77); 97i2a; 97i2b; 97i6c; 97v1a1 (91-94); 97v5a; 97v5d1; 98i3a; 98v1c (90-127); 98v2a; 98v5a; 100i3a; 100i5b; 101i5a; 101v1a; 103v2c (90-113); 108v2c; 109v2d; 112v3b (90-133); 114i2d; 119v3e; 120v2b (90-185); 128v2c (90-162); 131i2c (90-131).

Jan 90: 279v1a (91-173); 285i1a (90-207); 289i1a; 289i1c; 302i1b (91-191); 305v2a (91-139); 313v2d1; 313v2d2 (90-217); 314v2b; 332i3a (91-147); 336v2a (91-71); 336v2b; 338v1b (91-189).

Sep 90: 365v1a; 368va; 376ia (91-437); 377va; 389v2a; 408i1a; 414v1d; 414v2a; 414v5a; 415v5a; 417i1a (91-436); 417i2c; 417i2d 417v1a; 417v2a; 417v2b; 417v2d; 417v4a2; 417v5a; 418v3b; 421v2a; 421v5b; 423i1e; 423v3a (91-315); 424v3a; 424v4a; 426i2b; 426v2b (91-383); 427i2a; 427v3a; 428v1d; 428v2a (91-381); 428v3c; 429v1a (91-435); 429v2a; 435i1b; 435v5c; 435v6b; 436i1a (91-382); 436i3a (91-316); 436i3c; 436v4e (91-546); 438i3a (91-317); 439v3a; 439v6a; 441i3a; 441i4b (91-307); 442i4b; 442i6a; 442v1a; 442v3c; 442v5c; 444v2d; 447v1c (91-501); 449i1c; 453i2d; 454i1b (91-506); 466v1d; 478i2a; 478i2b; 478i2c; 479ic; 480i1b (91-314); 481va; 483v1b; 483v2a (91-434); 485v1b.

Hypoxylon stygium (Lév.) Sacc., Syll. Fung. 1: 379. 1882.
Anamorph: *Nodulisporium* (Plate XII, Figs. 1-2)

Colony 3 cm diam. in 5 days; felty to wooly, azonate to faintly zonate towards edge of colony; whitish first, then dark blonde (5D4), hair brown (5E4), to brown (6E8); margin entire; amber yellow (4B6) exudate in aerial mycelium; stromatic hyphae not seen; reverse olive green (3D7) to yellowish brown (5E8) at center; raw Sienna (6D7) pigment diffusing. Conidiophores abundantly produced throughout surface of colony, > 90 µm long x 2.7-3.6 µ m wide, sub-hyaline to pale brown, verrucose, several times branching.

Conidiogenous cells cylindrical, terminal, 13-20 x 2.7-3 µm, solitary or in whorls, with circular secession scars. Conidia obovoid to oblong-elliptical, (3-)3.6-5.4(-6.3) x (1.8-)2-3.2(-3.6) µm, one-celled, with flat basal abscission scar, hyaline to pale brown.

Total number of isolates: 106.

Representative isolates at ATCC: 89-35, 90-186, 91-516, 91-542.

COMMENTS: This endophyte was isolated both from trees and seedlings at both sites; although it was present in both leaf tissues (midrib, and intervein region), it was more commonly found in the midrib region, and recovered from all 3 leaf age classes.

My isolates agree with descriptions of *Hypoxylon stygium* given by Martin (1968). In addition, culture descriptions of *H. stygium* collected in French Guiana (NY: Samuels 2622, KFR 86-5, unpublished) agree well with my isolates.

RECORDED AS ENDOPHYTE: none.

OTHER SUBSTRATA: On wood, and bark; *Acacia*, *Castanopsis*, *Fagus*, *Fraxinus*, *Myrica*, *Olea*, Palmae, *Persea*, *Quercus*, *Scorodophleus*.

DISTRIBUTION: warm-temperate to tropical.

ISOLATES EXAMINED: **Jan 89**: 30v2a2 (90-01); 35v2b (90-13); 39v1a (90-11); 41v1b; 57v2d (89-68); 60i1b (89-67); 60v1e; 63i4b; 71i2c (91-50); 75i3e2; 75v2d2 (89-165); 148v2a (90-07); 158i1b (89-01); 174v1a (89-33); 176v2c1 (89-44); 190i1a (89-35); 216i2b (89-69).

Sep 89: 25i2b (90-126); 37v2a (91-102); 39i2a (90-87); 49v2c (90-186); 82v1c (90-85); 86i1b; 91v5b (90-123); 97i4b; 98v5e; 104v5c; 107v1a; 108v2b (91-101); 114v1b (91-36); 114v1d.

Jan 90: 274v2a (91-231); 293i2a; 296i1b; 300i2b (91-187); 302v1e (91-211); 302v2b (91-233); 304i1a (91-256); 305i1a; 318v2d (91-258); 329v1c (91-284); 335i1c (91-257); 335v1d (91-194); 335v5d (91-245); 335v6c (91-222); 336i1a (91-108); 336i2b (91-288); 336v1a; 336v1c (91-141); 338v3d; 339i3b (91-230); 340v3d; 342v5a; 345v1a; 346v6d (91-236); 347i1c (91-226); 347i4b (91-180).

Sep 90: 364v1d2 (91-511); 410v2c (91-452); 410v2d; 417i2e; 417v2e; 417v4a1; 420v2d; 421v2c; 421v4a (91-516); 423v4e (91-456); 424v3e; 426v3a; 427i1a (91-453); 427i3a; 427v2b (91-386); 427v2e; 427v3b; 427v3c; 428i1b; 428v1a; 429v1b (91-451); 433i1b; 435v1b; 435v3b (91-512); 435v5d (91-395); 436i6a; 438v3c; 439i1a; 439i5b; 439v2e (91-318); 441i4c; 441v2d (91-457); 441v5d; 442i2c; 442v4d (91-319); 442v6a2; 451i2c; 453v2a (91-513); 456v2a; 458i2d (91-454); 458v1b (91-455); 458v1d; 459id; 462v1b; 480v1c (91-515); 480v1d (91-514); 480v2a; 480v2d; 486va.

Hypoxylon sp.

Anamorph: *Virgariella* (Plate XIII, Figs. 1-2)

CMD: Colony 1 cm diam in 5 days; opaque, azonate; aerial mycelium scant, brownish green, margin even; stromatic hyphae not seen; reverse dark brown (6F7); pigment diffusing brown (6E8). Conidiophores scattered on colony surface, simple to dichotomously branched, 63-85.4 µm long x 2.1-2.8 µm wide, hyaline to pale brown, smooth to slightly verrucose.

Conidiogenous cells terminal and intercalary, 13.5-27 x 3.6-4.5 µm, with discoid secession scars. Conidia obovoid to ellipsoidal, (5-)6-8(-9) x (2.8-)3.2-4.5(-5) µm, one-celled, flat to circular basal abscission scar, hyaline to pale brown, smooth, developing in small clusters at apex, as well as singly at the side of conidiophores.

Total number of isolates: 5.

Representative isolate at ATCC: 89-39.

COMMENTS: This endophyte was isolated both from trees and seedlings, most frequently at high várzea. It was recovered from the midrib and intervein regions of the three leaf ages.

According to Petrini and Müller (1986) *Hypoxylon* species known to produce *Virgariella* anamorphs are *Hypoxylon fuscum* (Pers.:Fr.) Fr., *H. julianii* L. E. Petrini, and the *H. rubiginosum* (Pers.:Fr.) Fr. - complex. However, none of the species descriptions above mentioned matched my isolates.

ISOLATES EXAMINED: **Jan 89**: 47v3b (89-70); 179i1b (89-39); 179i1c; 186v2a.

Sep 89: none.

Jan 90: 282i6a.

Sep 90: none.

Nodulisporium sp. (Plate XIV, Figs. 1-3)

Colony 4 cm diam. in 7 days; velvety with radial strands, faintly zonate; first white, then darkening to mouse grey (5E3) from the center outwards; margin entire to minutely scalloped; dark brown, coiled stromatic hyphae (Fig. 3); reverse pale orange (5A3) to salmon (6A4), no pigment diffusing. Stromata formed after 1 month of incubation, in the periphery of colony; pulvinate, under 0.5 cm tall, first white then tan (6E6). Conidiophores forming in palisades over the surface of pulvinate stromata, to 54 µm long x 2.7-3.6 µm wide, hyaline to subhyaline, smooth, dichotomously branched. Conidiogenous cells terminal, cylindrical, 9-21.6 x 3.6-4.5 µm, hyaline, smooth, with round to conical secession scars. Conidia obovoid, (4.5-) 5.4-7.2 (-9) x (2.7-) 3-4.5 (-5.4) µm, one-celled, with flat basal abscission scar, subhyaline, smooth.

Total number of isolates: 19.

Representative isolates at ATCC: 90-117, 90-151.

COMMENTS: An endophyte isolated from both trees and seedlings at high and low várzea, but more abundant in seedlings, and at high várzea; occurring more frequently in the midribs of mature leaves.

ISOLATES EXAMINED: **Jan 89**: 86v2c; 98i2a.

Sep 89: 50v1a; 59v1a; 91v6e (90-117); 95v3b; 113v2a (90-151); 113v2b; 115i2d.

Jan 90: 280i1a (91-99); 280v1a (91-112); 293v1d (91-136); 296i1c; 310v3a (91-152); 332v3a (91-137); 341v1d (91-138).

Sep 90: 417v3a2; 436v5d (91-330); 452ib (91-329).

Penzigia* ? *indica Rawla & Narula, Indian Phytopath. 37: 312. 1984.
Anamorph: *Nodulisporium*. (Plate XV, Figs. 1-2)

Colony 2 cm diam in 5 days; velvety with scattered pale orange tufts developing later, azonate; orange white (6A2) then pale orange (6A3); margin even; colorless exudate in aerial mycelium; reverse orange grey (5B2) to golden blonde (5C4). Stromata produced at center of colony, cylindrical to capitate, to 1.8 cm tall, unbranched or apices with several digitate projections, salmon, then becoming blackish and villose at base. Conidiophores forming in palisades on upper surfaces of stromata, to 82.5 μm long x 3.3 μm wide, hyaline, smooth, dichotomously branching near base. Conidiogenous cells cylindrical, terminal, 13.5-25.2 x 2.7-3.6 μm, with round secession scars. Conidia ellipsoidal, (3.6-)5-6.3(-7) x (1.8-)2.7-3 (-3.6) μm, 0-septate, with flat basal abscission scar, hyaline, smooth.

Total number of isolates: 4.

Representative isolate at ATCC: 89-25.

COMMENTS: This endophyte was isolated from trees at high várzea, and occurred in the midribs of newly expanded and mature leaves.

Cultural and anamorphic characteristics of these isolates are much like *Penzigia* cf. *indica* Rawla & Narula described by Callan and Rogers (1990), except that the stromata produced by my isolates were salmon rather than blackish as described by those authors.

RECORDED AS ENDOPHYTE: none.

OTHER SUBSTRATA: on dead angiosperms.

DISTRIBUTION: tropical.

ISOLATES EXAMINED: **Jan 89**: 198v2b (89-25).

Sep 89: none.

Jan 90: 276v; 281v2c (91-114); 281v2d.

Sep 90: none.

Ustulina ? ***deusta*** (Hoffm.: Fr.) Petrak, Ann. Mycol. 19: 279. 1921.

Anamorph: *Nodulisporium* (Plate XIII, Figs. 3-4)

Colony reaching 1.5 cm diam. in 5 days; felty to appressed, scalloped zonation; yellowish greenish (1B8) to olive yellow (1C8); margin white, minutely plumose; a yellowish exudate forming in aerial mycelium; stromatic hyphae not seen; reverse peach (7A4); no pigment diffusing. Vertical compact hyphae formed after 1 week of incubation, abundant within zonation; pale to yellowish green at base, and pale brown towards apex, to 2 mm tall. Conidiophores compactly aggregated, arising along length of each vertical tuft in a palisade, 40-50 µm long x 4.5 µm wide, yellowish brown, smooth, unbranched to dichotmously branched near base. Conidiogenous cells terminal, cylindrical to clavate, with apical, circular secession scars. Conidia, oblong, (5.4-)6-7.5(-9) x (2.7-)3-4.5(-5.4) µm, one-celled, with flat basal abscission scar, hyaline to yellowish brown, smooth.

Total number of isolates: 36.

Representative isolates at ATCC: 91-157, 91-158, 91-298, 91-431.

COMMENTS: This endophyte was almost always present in seedlings, with its highest frequency in the high várzea. It was recovered most often from the midribs of both newly expanded and mature leaves.

The descriptions of *Ustulina deusta* given by Hawksworth (1972), and by Jong and Rogers (1972) [as *Hypoxylon deustum*] match my isolates.

RECORDED AS ENDOPHYTE: in *Abies alba* Miller, *Calluna vulgaris* (L.) Hull, *Erica carnea* L., *Nicotiana tabacum* L., *Vaccinium myrtillus* L., *Vaccinium vitis-idaea* L. (Petrini 1984b), *Triticum aestivum* (Riesen and Sieber 1985), *Ulex europaeus* L., *U. gallii* Planch (Fisher et al. 1986), *Chamaecyparis thyoides* (Bills and Polishook 1992).

OTHER SUBSTRATA: *Acer, Camellia, Citrus, Cocos, Coffea, Elaeis, Fagus, Hevea, Nicotiana, Theobroma, Tilia, Ulmus.*

DISTRIBUTION: cosmopolitan.

ISOLATES EXAMINED:**Jan 89**: none.

Sep 89: 51v2a; 60i2a; 65v2a; 69v2a; 76v2b; 86i1a; 90v2a2; 91i1c; 91v2a; 91v3b; 91v6a; 94i2a; 94v3a; 95v4a; 97v1a2; 100v1a; 100v2c; 100v4a; 101v3b; 103i6a; 103v1a; 103v3a; 103v5a; 103v5b; 103v6b; 104v6b.

Jan 90: 297v2b (91-106); 313v1e (90-216); 313v1e2; 314i3d (91-157); 338v3c (91-158); 346v6a; 346v6b.

Sep 90: 415v4b; 427v2c (91-431); 441v5a.

Xylaria adscendens (Fr.) Fr., Nova Acta Regiae Soc. Sci. Upsal. Ser. 3, 1: 128. 1851.

Anamorph: *Nodulisporium* (Plate XVI, Figs. 1-4)

Colony 4.5 cm diam in 7 days; velvety to appressed, eventually forming radial strands, with white, abundant cottony mycelium around point of inoculum, and reaching lid of Petri dish; faintly zonate; at first white, then covered by a layer of warty canary yellow (2B7) mycelium (Fig. 3), darkening to olive yellow (3D7), to blackish olive (2G6) from the center outwards; margin white to canary yellow, minutely plumose; dark brown, long stromatic hyphae with short protuberances (Fig. 4) in the aerial mycelium; reverse golden blonde (5C4); no pigment diffusing. Stromata formed after 1 week at point of inoculum or peripherally; cylindrical, robust, to 3.5 cm tall, 0.5 cm wide, eventually branching when touching Petri dish lid, first white orange to canary yellow then darkening to brownish grey at base with canary yellow to pale salmon at tip, villose at base; colorless to yellowish droplets forming along length of stromata. Conidiophores forming after three to four weeks, in palisades at tip of stromata, to 31.5 µm long x 3.6 µm wide, hyaline, smooth, branching near base. Conidiogenous cells, solitary, terminal, cylindrical, 13.5-22.5 x 3.6-4.5 µm, with denticulate secession scars. Conidia ellipsoidal, (8.1-)9.9-11.7 x 2.7-3.6 µm, one-celled, with flat basal abscission scar, hyaline, smooth.

Total number of isolates: 115.

Representative isolates at ATCC: 91-144, 91-334, 91-577, 91-583, 91-586.

COMMENTS: A widespread endophyte isolated from trees and seedlings at both high and low várzea, but more common at high várzea. It was recovered from the midrib and intervein regions of all three leaf-ages.

The colony morphology, stromal characters, and anamorph features of this endophyte agree well with those described by Rogers (1984a) and Callan and Rogers (1990) for *X. adscendens*.

RECORDED AS ENDOPHYTE: none.

OTHER SUBSTRATA: Indet. angiosperm, *Quercus*, *Sapindus*.

DISTRIBUTION: French Guiana, Hawaii, and Mexico.

ISOLATES EXAMINED: **Jan 89**: 30i2a; 30v1d; 35v1b (91-584); 35v2c; 37v2e1; 37v2e2; 41i2a; 41v2b; 49v4a (89-87); 57v1a1 (90-04); 57v1a2; 88v3b (89-84); 96v1b; 96v2a (89-88); 98v1a (89-91); 98v1b1; 209v1b; 211i1a; 218v1a; 218v1b; 218v1c; 218v1d; 218v1e; 218v2c2; 221i1b; 221i1c.

Sep 89: 29v1a; 49v1a; 51v1a; 58v2b; 59v1b; 78v2a; 91v6d; 94v1e; 98v5d; 100v4b; 103v6a; 103v6c; 103v6e; 104v5a; 108v1a; 109v1d; 123v1a.

Jan 90: 281v1a; 281v2a; 283v1c (91-79); 286v1a (91-80); 286v1b; 286v6a; 288v1c (91-160); 289v1b; 289v2d (91-54); 290v2e (90-220); 291i1a (91-144); 291v3a; 291v3c; 291v4b; 291v4e (91-143); 291v5c; 293v1a2; 293v1b; 293v2a (90-223); 293v2e; 294v1b; 294v2e; 295v2a; 296i1a (90-221); 296v1a; 296v1b (91-83); 296v2b (91-53); 297v2c (91-203); 298v2b; 298v2d (91-145); 300v2b; 301i2a (91-81); 301i2b; 301v1e (91-05); 302v1a; 303v1c (91-04); 304v1b (91-82); 304v2a; 308v3c; 314i2a; 314v2a; 314v3a (90-222); 315v1a; 315v1b; 317v2a; 319v3b; 324i3a (91-192); 325v3b (90-197); 329v1d; 330i2c (91-25); 330v2c; 334v4a; 334v4b; 335v1a; 335v2c; 336v3a (91-246); 336v5c; 340v2b (91-121); 347i4d.

Sep 90: 420v6a; 441v1b (91-361); 444i5b (91-384); 444v2a (91-334); 445v1e2 (91-409); 445v5a; 445v5e (91-322); 448va; 456i1b (91-363); 466v1c (91-385); 478v1c; 480v1a; 481ia.

Xylaria allantoidea (Berk.) Fr., Nova Acta Regiae Soc. Sci. Upsal. Ser. 3, 1: 127. 1851.

Anamorph: *Nodulisporium* (Plate XVII, Figs. 1-2)

Colony 2.5 cm diam in 5 days; cottony with radial hyphal strands, azonate to irregularly zonate; white to pale salmon then overlain with cocoa (6E6) mycelium; margin minutely plumose; stromatic hyphae not seen; yellowish exudate forming in the aerial mycelium; reverse greyish orange (6B5) to Sahara (6C5); no pigment diffusing. Stromata produced after 3 weeks at center, or at periphery of colonies; cylindrical, to 3.5 cm high, 3 mm wide, unbranched, at first pale orange (6A3) then sunburn (6D5). Conidiophores produced along the length of stromata as well as in tufts on surface of colony, forming in a palisade, to 72 µm long x 2.7-3.6 µm wide, sub-hyaline to pale brown, smooth, branching near base. Conidiogenous cells solitary, terminal, cylindrical, 31.5-45 x 3.6-4 µm, hyaline, with discoid to denticulate secession scars. Conidia clavate to ellipsoidal, few with a corona-like structure at apex, (6.3-)6.8-9.7(-10.8) x (3-)3.6-4.5 µm, one-celled, with flat basal abscission scar, hyaline [dust (5D2) in mass, smooth.

Total number of isolates: 15.

Representative isolates at ATCC: 89-16, 91-229, 91-441.

COMMENTS: This endophyte was isolated both from trees and seedlings, more often in the high várzea where it occurred in the midrib and intervein regions of the three leaf ages.

These cultures match the description of *X. allantoidea* given by Callan and Rogers (1990).

RECORDED AS ENDOPHYTE: none.

OTHER SUBSTRATA: on wood.

DISTRIBUTION: warm temperate and tropical.

ISOLATES EXAMINED: **Jan 89**: 75v3a (89-16); 186v2d (90-68); 230i1a; 230i1b.

Sep 89: 59v2c; 60i1a (91-442); 77v1a; 77v1b; 108v1c (91-441).

Jan 90: 310i1a (91-182); 334v6d (91-229).

Sep 90: 427v2a; 453v1e; 459ib; 480i2a (91-327).

Xylaria anisopleura (Mont.) Fr., Nova Acta Reg. Soc. Sci. Upsal. Ser. 3, 1: 127. 1851.

Anamorph: *Nodulisporium*. (Plate XVIII, Figs. 1-3)

Colony 1.5 cm diam in 5 days; velvety to appressed, occasionally whitish floccose sectors forming, azonate to faintly zonate; first white, then overlain with café-au-lait (6D3) to greyish brown (6E3) mycelium formed of subhyaline, coiled stromatic hyphae; margin plumose; surface of older cultures radially furrowed; reverse non pigmented or greyish orange (5B4). Stromata developing at edges of colony, and in more or less concentric zones, cylindrical, 0.4-1 cm tall, to 2 mm wide, unbranched, whitish, then tan, with orange to brownish orange exudate along the length. Conidiophores located at tip of stromata as well as on compact hyphal tufts forming on colony surface, hyaline to pale brown, smooth, irregularly branched near base. Conidiogenous cells solitary, terminal, cylindrical, 22.5-40.5 x 4.5-5.4 µm, with round to conical secession scars. Conidia ellipsoidal, (7-) 8-9 x 3.6-4.5 µm, one-celled, with flat basal abscission scar, hyaline.

Total number of isolates: 18.

Representative isolates at ATCC: 89-85, 89-157, 91-130, 91-282.

COMMENTS: An endophyte isolated from trees and seedlings both from low and high várzea. It was found more often in the midrib than intervein regions of newly expanded and mature leaves.

This endophyte matches the cultural description given by Callan (1988) for *X. anisopleura*. According to Rogers et al. (1987; 1988), *X. anisopleura* is a tropical variant of *X. polymorpha* (Pers.: Fr.) Grev.

RECORDED AS ENDOPHYTE: none.

OTHER SUBSTRATA: On bark and wood of trees including *Quercus* and palms.

DISTRIBUTION: tropical.

ISOLATES EXAMINED: **Jan 89**: 41v1c (89-12); 86v2b; 158i1a (89-85); 198v1a (89-119, 89-157); 209i2c.

Sep 89: 82v1b (91-52); 94v4d (91-51); 98v2d; 108v3a; 109v3a.

Jan 90: 285v2a; 296v2c (91-282); 310i2a (91-130).

Sep 90: 414v4d (91-460); 421v1d (91-561); 436v1b (91-459); 439v4b (91-499); 442v1c (91-500).

Xylaria arbuscula Sacc., Michelia 1: 249. 1878.

Anamorph: *Nodulisporium.* (Plate XVIII, Figs. 4-6)

Colony 3 cm diam in 7 days; velvety to appressed, faintly zonate; at first white, then darkening to cocoa (6E6) to burnt umber (6F6) but remaining white close to the point of inoculum; margin white, minutely plumose; stromatic hyphae long, with short, lateral protuberances (Fig. 6) in the aerial mycelium; colorless to yellowish exudate forming in the aerial mycelium; reverse pale orange (5A3) to brownish orange (6C8), eventually forming a dark brown ring at center. Stromata produced after 10 to 15 days in concentric zones, cylindrical, 1.5 to 3.5 cm high, to 2 mm wide, unbranched to 2-3 times branched towards apex, first white then darkening to dark brown to almost blackish with white apices, base villose with colorless exudate; or pulvinate, ca. 1 mm tall x 1 mm wide, white. Conidiophores formed in a palisade at apex of stroma, to 59.5 x 2.7-3.6 µm, smooth, branching near base. Conidiogenous cells solitary, terminal, cylindrical, 6-15 x 3.6-4.5 µm, with round secession scars. Conidia ellipsoidal, (4-)4.5- 6.3(-7) x 2.7-3.6 µm, one-celled, with flat basal abscission scar, hyaline, smooth.

Total number of isolates: 159.

Representative isolates at ATCC: 91-91, 91-167, 91-177, 91-183, 91-249, 91-306, 91-587.

COMMENTS: This endophyte was isolated in equal numbers from trees and seedlings at high várzea. It occurred more often in the midrib than in the intervein regions, and was isolated from newly expanded and mature leaves.

This endophyte fits well in the description of *X. arbuscula* given by Martin (1970) and Callan and Rogers (1990).

RECORDED AS ENDOPHYTE: none.

OTHER SUBSTRATA: on bark, and wood; indet angiosperm, and gymnosperm; *Cephalanthus*, *Quercus*, *Swietenia*.

DISTRIBUTION: warm temperate and tropical regions.

ISOLATES EXAMINED: **Jan 89**: 8va; 49v4a (89-087); 57v1a2; 88v3b (89-084); 98v2c; 103v1c; 110v1c; 110v2e; 158v1b; 221v2c; 221v2d (89-090).

Sep 89: 55i1a (89-107); 58i2a; 77i2a; 87v2a; 91v2b; 92i3a; 92v2a; 92v2b; 98v3b1; 106v1a; 112v3a; 114i2a.

Jan 90: 240v2b; 244v1b; 262v1a; 279i1a; 280i1b; 285v1a (91-163); 286i6a; 286v5a (91-90); 289i2a; 290v1d (91-261); 291v1a; 291v2b (90-237); 291v6b; 291v6c; 294v2d (90-235); 296v2a (91-162); 297v1a; 297v1b; 297v1c; 297v1d; 298v1e; 298v2c; 300i1a (91-002); 300i1b; 300v1e (91-091); 300v2d; 301i2c (91-003); 303i1d (91-183); 304v1a (91-166); 305v1b; 305v1c; 307i2a (91-204); 309v3c; 310v1a; 313v2c; 314i3a; 314v3e (90-239); 317v1c; 322i3b (91-275); 322v2b; 322v3a; 322v3b; 322v3e; 323v3a; 323v3b; 325v1c (90-234); 325v1d (90-238); 327i2b; 331i4d (91-125); 332v3d (91-167); 332v5d (91-248); 334v3d; 334v3e (91-127); 334v6a (91-206); 334v6b; 334v6c (91-181); 334v6e; 335v2b (91-249); 335v3d (91-207); 335v3e (91-289); 335v4a; 335v4b; 335v6a; 336v2d (91-239); 336v4b (91-208); 336v4d; 336v5a (91-210); 336v5b; 338v1a; 338v2a; 338v2b; 338v3b; 338v3e; 339v2a; 339v2b; 339v2c; 340i1b (91-177); 340v1a; 340v3a (91-

205); 347v2a; 347v2b; 347v3a; 347v3b; 347v5a (91-225); 347v5b; 347v5d (91-247); 347v6c (91-161).

Sep 90: 417v6d; 418i3c; 421v3c (91-368); 426i1a; 432i1a; 432v1a; 432v2b (91-461); 432v3a; 433i2a; 434i1a; 436v1a (91-373); 436v4c (91-306); 436v5a (91-443); 438i2a; 438v1d; 438v5a; 439i3b; 439i5a (91-343); 444i6a (91-304); 444i6b; 444i6c; 444v3a; 444v6b; 444v6c; 445i2c (91-389); 445i4a (91-426); 445i5b (91-444); 445v6e; 445v1e1; 450vb; 452i1a; 453v2b (91-372); 456v1a; 459vd; 465i1a; 465v1a (91-300); 465v2c; 465v2d; 467v1c; 467v1d2 (91-535); 467v2a; 478v1a (91-371); 478v2b (91-440); 483i1a (91-428).

Xylaria* ? *castorea Berk., in Hooker, Flora Novae-Zelandiae II. p. 204. 1855.

Colony 3.5 cm diam. in 5 days; velvety to appressed, azonate; pale orange (5A3) to pale salmon, then darkening to brownish orange (6C3) towards edge; margin entire; stromatic hyphae not seen; reverse non pigmented. Stromata produced after 15 days, scattered, cylindrical, with broad bases and acute apices, to 3 cm tall, 3 mm wide, unbranched, pale salmon then darkening to brown from the base upwards, villose. No conidiogenous cells observed.

Total number of isolates: 10.

Representative isolates at ATCC: 89-152, 91-252, 91-331.

COMMENTS: An endophyte isolated more often from trees, at both low and high várzea; occurring in the midribs as well as in the intervein region of newly expanded and mature leaves.

Cultural characteristics described by Martin (1970) and Rogers and Samuels (1986) are very similar to those of my isolates. Their cultures also remained sterile.

RECORDED AS ENDOPHYTE: none.

SUBSTRATA: On wood of unrelated angiosperms, including *Beilschmiedia*, *Fagus*, *Griselinia*, *Nothofagus*, *Pittosporum*, *Pseudopanax*, *Weinmannia*; and gymnosperms, including *Dacryocarpus*, *Phyllocladus*, *Podocarpus*. Specimens recorded (Martin 1970) from South Africa and Brazil were from unidentified hosts.

DISTRIBUTION: tropical.

ISOLATES EXAMINED: **Jan 89**: 1i2a (89-150); 1v3a; 1v6a; 90v3d.

Sep 89: none.

Jan 90: 283v2a (91-252); 295i1c; 347i3a.

Sep 90: 364v1a (91-525); 421v3b (91-524); 465v1e (91-331).

Xylaria coccophora Mont., Ann. Sci. Nat. Bot. Ser. 4, 3: 109. 1855.

Anamorph: *Nodulisporium* (Plate XIX, Figs. 1-4)

Colony 2 cm diam in 5 days; velvety, zonate; at first white, then covered by a layer of warty canary yellow (2B7) hyphae (Fig. 4), finally darkening to greyish yellow (3C4), to olive yellow from the center outwards, with pale yellow (3A3) zonation; margin pale yellow (3A3), plumose; dark brown, short stromatic hyphae formed in the aerial mycelium (Fig. 3); reverse non pigmented to orange grey (5B2); no pigment diffusing. Stromata produced in concentric zones, narrowly cylindrical, to 2 cm tall, 1 mm wide, unbranched, at first white, then blackish, wrapped in canary yellow (2B7) hyphae, with a colorless exudate at base of stromata. Conidiophores forming in palisades over surface of stromata and in zonations on surface of colony, branching near base. Conidiogenous cells solitary, terminal, cylindrical, 18-27 x 4.5-6.3 μm with denticulate secession scars. Conidia ellipsoidal, (7.2-)8-9 x 2.7-3.6 μm, one-celled, with flat basal abscission scar, hyaline, smooth.

Total number of isolates: 15.

Representative isolates at ATCC: 91-171, 91-174, 91-260.

COMMENTS: This endophyte was isolated from trees and seedlings at high and low várzea. It was primarily isolated from the midrib regions of newly expanded and mature leaves.

These endophyte cultures agree well with the description given by Callan (1988) and Rogers et al. (1988) for *X. coccophora*.

RECORDED AS ENDOPHYTE: none.

OTHER SUBSTRATA: on wood.

DISTRIBUTION: neotropical.

ISOLATES EXAMINED: **Jan 89**: 149i1e; 176v1b; 176v1c.

Sep 89: 94v6b; 97v5c.

Jan 90: 302v2e (91-174); 303i2b (91-171); 305v2b (91-260); 318v2a; 336v2c; 340i2a (91-238).

Sep 90: 417v1b; 417v3d; 423v3c; 436v6a1 (91-540); 484va.

Xylaria cubensis (Mont.) Fr., Nova Acta Reg. Soc. Sci. Upsal. Ser. 3, 1: 126. 1851.

Anamorph: *Xylocoremium flabelliforme* (Schw.: Fr.) J. D. Rogers, Mycologia 76: 914. 1984. (Plate XX, Figs. 1-2)

Colony 4 cm diam in 5 days, cottony with appressed sectors, azonate to faintly zonate; pale orange (6A3) to salmon (6A4); margin white, minutely plumose to even; exudate colorless forming in the aerial mycelium; stromatic hyphae in the aerial mycelium lacking; reverse non-pigmented to pale orange (5A3). Stromata forming after 5 days, first located in the center then abundantly over colony; cylindrical or flabelliform, to 2.5 cm tall, occasionally villose at base, pale salmon, eventually darkening to brown at base. Conidiophores in palisades, developing abundantly on upper part of stromal surface, appearing floccose; to 36 µm long x 3 µm wide, hyaline, smooth, several times branching. Conidiogenous cells solitary, terminal, cylindrical, 9-18 x 2.7-3.6 µm, with round to denticulate secession scars. Conidia obovoid to ellipsoidal, (3.6-)5-6.3 x 1.8-3.6 µm, one-celled, with flat basal abscission scar, hyaline, smooth.

Total number of isolates: 673.

Representative isolates at ATCC: 89-151, 91-601, 91-602, 91-603, 91-604, 91-605, 91-606, 91-607, 91-608, 91-609, 91-610, 91-611, 91-612, 91-613, 91-614, 91-615, 91-616, 91-617, 91-618, 91-619, 91-620, 91-621, 91-622, 91-623.

COMMENTS: This endophyte was the most dominant species of the endophytic leaf community in *E.oleracea*. It was isolated almost equally from trees and seedlings at high várzea, and was more frequently found in seedlings than trees at low várzea. *Xylaria cubensis* was very common in the midrib regions of the three leaf age classes.

The endophyte colonies fit well with published descriptions of *X. cubensis* (Jong 1970; Martin 1970; Rogers 1984b; Rogers and Samuels 1986; Nagasawa 1988; Callan and Rogers 1990). However, *X. cubensis* is a morphologically variable species, indicating that it is a complex comprising more than one taxon. Resolution of this complex will require morphological and biochemical studies of more cultures from ascospores than have been done so far.

RECORDED AS ENDOPHYTE: in *Licuala ramsayi* (Rodrigues and Samuels 1990), *Chamaecyparis thyoides* (Bills and Polishook 1992).

OTHER SUBSTRATA: On wood of *Acer*, *Camellia*, *Magnolia*, *Protium*, and *Quercus*.

DISTRIBUTION: cosmopolitan.

ISOLATES EXAMINED: Jan 89: 1i3b (89-05); 1i4b; 1v4a; 1v4b; 1v4c; 1v5a; 1v6b; 6va; 10ia; 10vb (89-06); 30i1b; 33i1a; 33i1b; 33v2e; 35v1c; 37v1b2; 37v2d2 (89-151); 39v2e; 41v2c; 57i3b; 57v3a; 60v1b; 60v3c; 63i4a; 69i1c; 69v3d; 71v2a; 73v1a; 84i3a; 84v1a; 84v3b; 86v2a; 86v2e1; 86v3a; 88v2b; 90i1b; 90i2a; 90v1a; 90v1b; 90v1c; 90v2c; 96v1a; 100i2d; 100v1b; 103v1b; 107i1b; 107i2b; 107v2a; 110ia; 110i2c; 110v2d; 114v2a;

141v2a, 153v1a; 156v1b; 160i2a; 169i2d; 169v2c; 171i2a; 171v1a; 171v1b; 174i1b; 174i2d2; 174v2b1; 176ib2; 176i1b; 176i2c; 176v2a; 176v2b; 176v2c2; 184v2a; 192i2b1; 192i2c; 192v1a; 198v1c; 200i1a; 200v1a; 209i1a; 209v1b; 209v1c; 209v2b; 209v2c; 209v2d; 214i1a; 214v1b; 214v1c; 214v2a; 214v2b; 214v2c; 214v2d; 216v1c; 216v2d; 218v2a; 218v2b1; 218v2c; 218v2d; 218v2e; 223i2a2; 223i2b; 225v2c; 228v1a.

Sep 89: 15v1a; 48v1a; 49v1c; 50v2c; 50v2d; 50i1b; 51v1b; 58v1c; 58v2a; 58v2c; 59i2a; 59i2c; 59i2d; 59v1c; 59vd; 59v2a; 59i1c; 59i1b; 60v1b 60v1c; 60v1d; 60v2a; 60v2c; 67i2a; 67i2b; 67v1b; 67v1c (91-95); 67v2a; 67v2c; 67v2d; 68v1a; 68v2a2; 68v2b; 68v2d; 69i1b; 69v1b; 74v1a; 75v2a; 76i2a; 76v2a; 77v1e; 77v2a; 77v2e; 77v2c; 77v2d; 77v1c; 77v2b; 78i1b; 78v2b; 78v2c; 82v1a; 82v1d; 85i1b; 85i1c; 85i2a; 85v1a; 85v1b; 85v1d; 86v1a; 86v1b; 86v1c; 86v1e; 86v1d; 86v2a; 86v2c; 86v2e; 87v1b; 87v1d; 87v1a; 87v1b; 87v1c; 87v2c; 87v2b; 91v2d; 91v6b; 91i3a; 91i4a (90-182); 91v1a; 91v1b (90-177); 91v2c; 91v4a; 92v3a; 92v3c; 92v4a; 92v4d; 92v5c; 92v5e; 92v6b; 92i1b; 92i4b; 92v5d; 92v6c; 92i6a; 94v1d; 94v1c; 95v4c; 95v4b; 95v4e; 97v2d; 97v1b; 97v6b; 97v1c; 97v2a; 97v2b; 97v3a; 97v3c; 97v4b; 97v4e; 97v5e; 97v6a; 97v6c; 97v2c; 97v5d2; 97v1d; 97v6d; 98v2b; 98v4b; 98v4c; 98v4e; 98v5b; 98v6b; 98i4a; 100i4b; 100i5a; 100v1b; 101i6a; 103v4a (90-178); 103v6c, 103v2b; 104v6c; 108v2a; 109v2a (90-180); 119v1b; 121v3d (91-581); 123v1c; 129v3d.

Jan 90: 238v1a; 238v2a; 240v2a; 249v2a2; 254i2a; 260v1a; 260v2b; 260v2c; 261v1d; 261v2a; 269i1a; 269i1b; 278v1a2; 279v1b; 280i2a; 280v1b; 281v1e; 281v2b; 283i1a; 283v1a; 283v1b; 283v1d; 285i1b; 286i2a; 286v5b; 286v5c; 286v6c; 287i5a; 288v1a; 288v1b; 288v2b; 288v2c; 289v1c;

289v1e (91-115); 289v2c; 290v1a; 290v1b; 290v1c; 291i1c; 291i2a; 291i3a; 291i3b; 291i5a; 291i6b; 291v1b; 291v2c; 291v3; 291v3d; 291v5a; 291v5b; 293i1b; 293i2c; 293v2c; 294v1c; 294v2c; 295i2b; 295v1b; 295v2b; 295v2e; 296i2a (91-104); 298i2b; 298v2a1; 298v2a2; 300v1c; 300v1d; 302v1c; 302v1d; 302v2d (90-189); 303i2a; 303v1d; 303v2c; 303v2e; 307i1a; 307v1c (91-603); 307v2b; 307v2c; 307v2d (91-615); 308i1a (91-616); 308v1c (91-597); 308v2b; 308v2c; 308v3a (91-606); 308v3b; 309i3b (91-621); 309v1b; 309v2c (91-620); 309v2e (91-605); 310v2d (91-283); 310v3b; 310v3c (91-599); 310v3d; 312v3a; 312v3b; 313i3a; 313v1c; 313v2a; 313v2e (90-219); 314i3c (91-234); 314v1a; 314v1b; 314v3c; 314v3d (90-190); 315i3b; 315v1c; 315v1d; 315v3c; 317v2b; 317v3b; 322v1c; 323i1c; 323i2c; 325v1a; 325v1b; 327i2c; 327v2a; 327v2b; 328v2a (91-172); 328v2b; 329i3e; 330v1d; 330v1e; 331i1b (90-191, 91-618); 331i1e (91-617); 331i2c (91-600); 331i3b (91-612); 331i3c; 331i5b (91-601); 332i1a (91-619); 332i2c (91-103); 332i3c; 332i4a (91-113, 91-598); 332i5a; 332v2d (91-151, 91-604); 332v3c; 332v4b; 332v5a; 332v6b (91-32, 91-614); 332v6c; 332v6d; 332v6e; 334v1c; 334v1d (91-105); 334v2b (91-66); 334v2d (90-236); 334v2e; 334v4c; 335i2e; 335i6a; 335v1b; 335v1c; 335v5b; 336i2c; 336v2e; 336v5e (91-267); 338i2c; 338i3a; 338v1c; 339i2c; 339v1a; 339v1c; 339v2d; 339v2e; 339v3b; 339v3c; 339v3e; 340i1c; 340i2b (91-149); 340i2d; 340i3b; 340i3c; 340v1e; 340v2d; 341v2a; 341v2b; 341v2c; 341v3e; 342v3a; 343i4d; 343v3a (91-24); 343v3d; 343v4a (91-33); 346ic; 346i2a; 346i2b; 346i4a; 346i4d (91-150); 346v1b; 346v2d; 346v2e1; 346v3d; 346v4a; 346v4c; 346v4d; 346v5a; 346v5b; 346v5c; 346v5d; 346v5e; 347i4a; 347i6c; 347i6d (91-200); 347v1c; 347v2e; 347v5e.

Sep 90: 363va; 364v2c; 367va; 394va; 395va; 397v2a; 404v2a (91-375); 407va; 414v5b; 414v6a; 417i4a; 417v2c; 417v4b; 417v4d; 417v4e; 417v5c; 417v6b; 418i1a; 418i2e; 418i6e; 420v2b; 420v3a; 423v2c; 423v2e; 423v3b; 423v4a; 423v4b; 423v4c; 423v4d; 426v1b; 426v2c; 427v1b; 427v1e; 432i1b; 432i2b; 432i2c; 432i5c; 432i6b; 432i6d; 432v4c; 432v5a; 432v5c; 432v5d; 432v5e; 432v6a; 432v6b; 432v6d; 433i2c; 433v3b; 435i4c (91-295); 435i5a; 435i5c; 435v3a; 435v4b; 435v4c; 435v5a; 436i3b; 436v2d (91-533); 436v4a; 436v4b; 436v6b; 436v6c; 438i3b; 438i4b; 438v1a; 438v2b; 438v3b; 438v3d; 438v3e; 438v4d; 438v4e; 439v3c; 439v5a; 439v5b; 441i1b; 441i2a; 441i2b; 441i3b; 441i6a; 441va; 441vc (91-390); 441v1c; 441v2b; 441v2c2; 441v2e; 441v3a; 441v3b1; 441v3b2; 441v4a; 441v4a2; 441v4b; 441v5b; 442i1a; 442i3b; 442i4c; 442v2b; 442v2c; 442v3b (91-321); 442v4b; 442v5a; 442v5b; 442v5d; 442v5e; 442v6b; 442v6c (91-376); 444i3a; 444i4d1; 444v3e; 444v4c; 445i1b; 445i4c; 445i4d; 445i6b; 445v1c2; 445v1d; 445v2c; 445v2d; 445v2e; 445v3c; 445v3e; 445v4d; 445v4e; 445v5b; 447i2b (91-623); 447v1a (91-320, 91-622); 447v1b (91-602); 447v2b; 447v2c; 447v2d; 448vb; 449v1c; 449v2d (91-410); 450ia; 450ic (91-607); 450vd; 451i2a; 451i2b (91-293, 91-610); 451i2d (91-609); 451v2a; 451v2d (91-611); 453i1a; 453i1b; 453i2a; 453v1a (91-613); 453v1c; 453v2c; 454i1a; 454va (91-608); 455i1a; 456v2b; 458i1b; 458i1c (91-374); 458i1d; 458i2e; 458v1c; 459ic; 459vb; 462v1a; 463va; 465v1c; 465v1d; 466ia; 466v1b; 467v2e; 468vd; 468ve; 478v2d; 478v2e; 479ib; 479vc; 479vd; 480v1e; 480v2d (91-294); 481ib; 485v2a.

Xylaria curta Fr., Nova Acta Regiae Soc. Sci. Upsal. Ser. 3, 1: 126. 1851.
Anamorph: *Nodulisporium* (Plate XXI, Figs. 1-3)

Colony 3.5 cm diam in 7 days; velvety to appressed; becoming overlain by feathery, white radial hyphal strands, azonate; at first white, then darkening to mouse grey (5E3) to elephant skin (5E2) from the center outwards; margin white, highly plumose, later rising from colony surface after touching the Petri dish edge and then developing into stroma-like structures; colorless exudate forming in the colony surface; dark brown stromatic hyphae forming in the aerial mycelium (Fig. 3); reverse non pigmented to orange white (5A2); no pigment diffusing. Stromata produced after 2 weeks at center of colony along rays or at periphery of colony; cylindrical, to 3 cm high x 2 mm diam, fragile, unbranched, to branched when touching Petri dish lid, at first white, then darkening to brownish grey to blackish, with white to white orange apices, base villose, with a colorless exudate. Conidiophores forming a compact palisade along the length of stromata after 3 weeks of incubation, to 72 x 4.5 µm, subhyaline, smooth, sparingly branched. Conidiogenous cells solitary, terminal, cylindrical, 40-55 x 4-5.4 µm, with denticulate conidial secession scars. Conidia, obovate to ellipsoidal, (6.3-)6.7-8.5(-9) x (2.7-)3.5-4.3(-4.5) µm, one-celled, with a flat basal abscission scar, hyaline, smooth.

Total number of isolates: 122.

Representative isolates at ATCC: 91-124, 91-299, 91-301, 91-303, 91-305, 91-333, 91-411, 91-412.

COMMENTS: An endophyte isolated from trees and seedlings located at high várzea. It was recovered in higher numbers from midrib regions of newly expanded and mature leaves.

The colony morphology and conidial size agree with descriptions given by Martin (1970), Rogers (1983), and Callan and Rogers (1990) for *X. curta*.

RECORDED AS ENDOPHYTE: in *Licuala ramsayi* [as *Xylaria* sp.] (Rodrigues and Samuels 1990), *Chamaecyparis thyoides* (Bills and Polishook 1992).

OTHER SUBSTRATA: on wood; *Acacia*, *Aleurites*, *Betula*, *Fagus*, *Malus*, *Tsuga*.

DISTRIBUTION: cosmopolitan.

ISOLATES EXAMINED: **Jan 89**: 30v2d.

Sep 89: 50i1a; 91v5a; 94v3c; 104v1a (90-78); 130v3b.

Jan 90: 283v2b (91-184); 286i3a; 298v1c (91-250); 314v1d 91-124); 335v2d; 336i3a (91-280); 336i3b; 347v1d (91-259).

Sep 90: 370vb; 406v1a (91-498); 427v1d (91-299); 433i1c (91-414); 433i2b; 433v3a; 433v4a; 433v5a (91-415); 433v6a (91-417); 437v1a; 439i3a; 439i5a; 439i6a; 439v2c (91-418); 439v2d (91-352); 439v4a (91-388); 439v4c; 441i1a; 441v1a; 441v5c (91-301); 442i3a (91-425); 442i4a (91-405); 442i5a (91-333); 442v1b; 442v4c (91-448); 444i2a; 444i3b (91-406); 444i3c; 444i4a; 444i4b; 444i4c; 444i4d2; 444i6d1 (91-303); 444v2b; 444v2c (91-419); 444v3c; 444v3d; 444v4a; 444v4b; 444v4d (91-449); 444v4e; 444v5a1; 444v5b; 444v5c; 444v5d (91-416); 444v6a; 444v6e (91-412); 445i2a; 445i2b; 445i2d2; 445i3a; 445i3b; 445i3c; 445i3d; 445i3e; 445i5a;

445i5c (91-507); 445i5d; 445i6a; 445i6c; 445i6d; 445v1a; 445v1b; 445v2a; 445v2b; 445v3a; 445v3b; 445v3d2; 445v4a; 445v4b; 445v5c; 445v5d; 445v6a; 445v6b; 445v6c; 447i2a (91-450); 447i2c; 447v2b (91-424); 449i1a (91-447); 449i1b; 449i1d (91-446); 449i2a (91-387); 449v1a; 449v1b; 449v1d; 449v1e; 449v2a; 449v2b; 449v2c (91-445); 450v1a; 456i1a;456i2b (91-305); 457ia (91-302); 457ib; 457va; 458i2a (91-505); 458i2c; 458v1a; 458v2a; 458v2c; 459vc; 459ve (91-411); 465v2b; 466v1a (91-413); 467v1b (91-421); 467v1d1; 468v1a; 468v1b.

Xylaria* ? *microceras (Mont.) Fr., Nova Acta Regiae Soc. Upsal. Ser. 3, 1: 128. 1851.

Colony 2.5 cm diam in 5 days; cottony with radial strands, faintly zonate; white to orange white (5A2), darkening to birch grey (5C2) at center of colony; margin plumose; stromatic hyphae not formed; reverse greyish orange (6B5); no pigment diffusing. Neither stromata nor conidiogenous cells produced.

Total number of isolates: 7.

Representative isolates at ATCC: 89-27, 91-427.

COMMENTS: An endophyte isolated mainly from seedlings, and restricted to low várzea; occurring in the midribs of newly expanded and mature leaves.

These endophytic isolates agree well with ascospore cultures of *X. microceras* originating in Taiwan, Puerto Rico, and Venezuela growing at

WSU lab. The only published descriptions of *X. microceras* were based on collections from Venezuela (Rogers et al. 1988), and from Puerto Rico and USA (Callan and Rogers 1990). Cultures from the teleomorphs collected in localities other than Puerto Rico and USA, did not produce stromata and remained sterile.

RECORDED AS ENDOPHYTE: none.

OTHER SUBSTRATA: on wood.

DISTRIBUTION: tropical.

ISOLATES EXAMINED: **Jan 89**: 71v4a (89-27); 71v4b; 71v4c; 71v4d.

Sep 89: none.

Jan 90: none.

Sep 90: 395vb (91-398); 426v2d (91-463); 428v1b (91-427).

Xylaria* ? *multiplex (Kunze) Fr., Nova Acta Regiae Soc. Sci. Upsal. Ser.3, 1: 127. 1851.

Colony 4.5 cm diam in 7 days; velvety to felty, faintly zonate with white, thin radial hyphal strands; at first white to orange white, then darkening to elephant skin (5E2) to beaver (5F4); margin white, thinly plumose; colorless to yellowish exudate produced in the colony surface; stromatic hyphae with short protuberances forming in the aerial mycelium; reverse beige (4C3); no pigment diffusing. Stromata rarely forming after 3 weeks, in the center of colony, in a more or less concentric pattern; cylindrical, robust, to 20 mm tall x 3 mm wide, unbranched to occasionally forked, first white then turning dark brown to almost black, except white at tip, tomentose at the base, with a yellowish exudate. Conidiogenous cells and conidia not observed.

Total number of isolates: 60.

Representative isolates at ATCC: 90-98, 91-109, 91-165, 91-401.

COMMENTS: This endophyte was isolated almost equally from trees and seedlings at low and high várzea. It occurs in midrib and intervein regions of newly expanded and mature leaves; only one isolate was recovered from an unopened leaf.

The stromatic hyphae produced could not be distinguished from those of *X. adscendens* and *X. obovata.*

Identification was made possible by comparison with cultures of *X. multiplex* derived from teleomorphs collected in Taiwan and South America growing at WSU. Martin's (1970, Plate IV, Fig. 6) photograph of *X. multiplex* taken from a culture is strikingly similar to my isolates. Martin's isolates and those growing at WSU also remained sterile.

RECORDED AS ENDOPHYTE: none.

OTHER SUBSTRATA: On wood; *Aleurites*, *Quercus*, and indet. angiosperm.

DISTRIBUTION: subtropical and tropical.

ISOLATES EXAMINED: **Jan 89**: 1v6c (89-144); 1v6d; 8vb (89-123); 10ib; 37i2c; 39v2c; 39v2d; 90v3b (90-14); 110v1c; 133v1a (89-116); 140v2b; 171i1b; 171i1c; 184i2b (89-121).

Sep 89: 50v2a; 54i2a; 95v5a; 98v1d (90-98); 100v2a.

Jan 90: 260v2a; 261v1c (91-178); 286i1a; 286i1b (91-255); 286i3b; 286v3a; 291v2a (91-198); 293v1a1; 293v1c; 297v2d; 304i2a (90-202); 315v3a (91-227); 322i3a (91-235); 323v1a (90-213); 323v1d; 323v2a; 331i2b; 335v5a (91-123); 335v6b; 336i4a (91-186); 336v3b (91-228); 339i2b (91-131); 339i2d (90-212); 340i2c; 344i4a; 344i4b; 346v2a (91-164).
Sep 90: 418v5a (91-401); 427v1a (91-467); 433v2a (91-508); 435i1a; 435i2a (91-351); 436i2a (91-429); 436v5b (91-402); 438i2a (91-339); 438v4a; 444v6d (91-536); 451i2a; 478v1b; 478v1e (91-370); 480v1b (91-403).

Xylaria obovata (Berk.) Fr., Nova Acta Regiae Soc. Sci. Upsal. Ser. 3, 1: 127. 1851.
Anamorph: *Nodulisporium* (Plate XXII, Figs. 1-3)

Colony 4 cm diam in 7 days; velvety to appressed, faintly zonate at center of colony; at first white then overlain with dark brown (6F7) to negro (6F3) layer of mycelium; margin white, plumose; exudate not produced; stromatic hyphae dark brown (Fig. 2); reverse brownish orange (6C8). Stromata formed at periphery of colony after 2 weeks, cylindrical to clavate, fragile, to 25 mm tall x 2-5 mm diam, unbranched, initially white, then turning dark brown; colorless exudate at base of stromata. Conidiophores forming in a palisade at tip of stroma, 45-54 µm long x 2.7-3.6 µm wide, subhyaline, dichotomously branching near base. Conidiogenous cells solitary, terminal, cylindrical, 18-27 x 3.6-4.5 µm, with conical secession scars. Conidia ovoid to obclavate, (8.4-)9.4-12 x 2.8-3.5 µm, one-celled, with flat basal abscission scar, hyaline, smooth.

Total number of isolates: 9.

Representative isolates at ATCC: 91-465, 91-469, 91-504, 91-545.

COMMENTS: This endophyte was present in seedlings and trees, but most commonly isolated from seedlings at low and high várzea. It was isolated almost exclusively from the midrib regions of newly expanded and mature leaves.

These isolates agree with the description of *X. obovata* given by Callan and Rogers (1990).

RECORDED AS ENDOPHYTE: none.

OTHER SUBSTRATA: on wood.

DISTRIBUTION: Puerto Rico, Venezuela.

ISOLATES EXAMINED: **Jan 89**: none.

Sep 89: 7v1a.

Jan 90: none.

Sep 90: 364v1b (91-466); 415v2a; 415v3b (91-465); 426v1a (91-408); 435v4a (91-332); 438v2a (91-469); 442i2a (91-545); 467v1a.

Xylaria* ? *palmicola Winter, Grevillea 15: 89. 1887.

Colony 3.5 cm diam in 7 days; velvety to appressed, faintly zonate; white first, then darkening to old silver (4E2) to blackish, becoming overlain with a greenish mycelium in a concentric pattern; margin minutely plumose, white; exudate not produced; reverse greyish orange (5B4). Stromata produced after 10 days, in concentric zones; cylindrical, 10 (-25) mm tall, robust with acute, white apex, erect to less frequently curved, unbranched, villose at base; first white then greyish beige (4C2) to dark grey (1F1), becoming clothed in olive green (3D7) hyphae, forming in more or less regular rings around stromata. Conidiogenous cells and conidia not observed.

Total number of isolates: 2.

Representative isolates at ATCC: 91-88, 91-340.

COMMENTS: This was isolated twice, from trees at high várzea. It occurred both in midrib and intervein regions of mature leaves.

Rogers and Samuels (1986) described similar cultural characters for New Zealand collections of *X. palmicola*, where the presence of a "blue ring around the centre of colony" and stromata "clothed in green hyphae" were noted. Conidia formed along stromata in their cultures.

RECORDED AS ENDOPHYTE: none.

OTHER SUBSTRATA: on *Euterpe* sp. (Viégas 1961), and on seeds of the palm *Rhopalostylis sapida* (Wendl.) Drude [Rogers and Samuels 1986].

DISTRIBUTION: Brazil, New Zealand.

ISOLATES EXAMINED: **Jan 89**: none.

Sep 89: none.

Jan 90: 297i1a (91-88).

Sep 90: 485v1c.

Xylaria* ? *telfairii Berk. & Fr., Nova Acta Regiae Soc. Sci. Upsal. Ser. 3, 1: 127. 1851.

Colony 3 cm diam in 5 days; velvety to appressed, faintly zonate; at first white, then becoming gray-brownish; coiled hyphae forming at surface of colony; colorless exudate forming in the aerial mycelium; margin minutely lobed, raised, developing into stromatal structures after 1 week; reverse non pigmented to pale orange (5A3); no pigment diffusing. Stromata formed at periphery of colony, fan-like, to 0.5 cm high, white to orange white to greyish orange (6B5). Conidogenous cells not observed.

Total number of isolates: 25.

Representative isolates at ATCC: 90-192, 90-232, 91-142, 91-310.

COMMENTS: An endophyte isolated almost equally both from seedlings and trees, at high and low várzea. It was found in the midrib and intervein regions, but was much more frequent in the former. It was recovered from the three leaf age classes.

Identification of these isolates was possible only after comparison with cultures of *X.telfairii* derived from teleomorphs that originated in South America that were growing at WSU, and which agree with the description given by Callan and Rogers (1990) for cultures from a specimen collected in French Guiana.

RECORDED AS ENDOPHYTE: none.

OTHER SUBSTRATA: on wood.

DISTRIBUTION: tropical.

ISOLATES EXAMINED: **Jan 89**: 174i1b1; 174i2b (89-26); 209i2a2 (89-109; 89-158); 214i1b1.

Sep 89: 49i2a; 55v1a; 67v1a; 92v2c; 94v4c (89-98); 98v6c; 100v3b.

Jan 90: 263v2e (91-159); 291v4c (91-153); 295v2d (91-107); 301i2e (90-232); 304i2c (90-192); 313v1a (90-193); 332v1a (91-170); 332v1b; 334v5a (91-142); 334v5b; 334v5e; 346v (90-201).

Sep 90: 436v2b (91-430).

Xylariaceae I

Colony 3 cm diam in 5 days; cottony to velvety, irregularly zonate; at first white to orange white (6A2), becoming overlain with canary yellow (2B7) hyphae with warty excresences in the center of colony; margin plumose; reverse non pigmented to brownish orange; no pigment diffusing. Neither stromata nor conidiogenous cells produced.

Total number of isolates: 94.

COMMENTS: The canary yellow pigmented hyphae with warty excresences are distinctive but not diagnostic. Similar hyphae have been reported by Rodrigues and Samuels (1989) for cultures of *Phylacia globosa* Lév. and *P. bomba* (Mont.) Pat. var. *macrospora* K. F. Rodrigues & Samuels, and by Callan (1988) for cultures of *Xylaria coccophora*. These ornamented hyphae have also been found in colonies of endophytic *X. adscendens* (see above).

ISOLATES EXAMINED: **Jan 89**: 30i2b; 57v2a; 60i1a; 63i2a; 63i4e1; 63i4e3; 86i3a1; 103i1e; 122v1a; 190i1b (90-71); 221v1e.
SEP 89: 49v1b; 58i1b; 77i2b; 85v2b; 85v2c; 94v5a; 94v5c; 97v3b; 98v6a; 100v6b; 103i4a; 103i4b; 108v3c; 114v2a; 114v2c; 115v1a; 115v2a; 115v3a; 119v3b; 121v1a; 123v3a; 125i2a; 125v1a; 125v1c; 125v3a (90-169).
Jan 90: 277v1; 278v1a1; 286v4b (91-154); 297v1e (91-85); 300v2e (91-67); 303v1a; 313i2a (91-156); 313i3b (91-55); 317v3a (91-120); 325v2e (90-198); 328v3a; 328v3b; 329v3c (91-119); 330v2d (91-56); 330v3e; 332v3b; 336i2a; 344v1a (91-56).
Sep 90: 408va; 410i1b; 414i2a; 414v4a; 414v4c; 414v5c; 415v3a; 417ib; 417i1c (91-433); 417i3a; 417i5a; 417v1c2 (91-313); 417v1d; 417v2b; 417v3b; 417v3e; 418i1d; 418i2a; 418i4b; 418i4d; 418i6a; 418i6c; 418v2a; 418v3c (91-379); 418v5b; 418v5d; 420v2a; 421v1c (91-312); 423i4a; 423v2d (91-311); 423v5a; 428v2b; 433v5b; 435v2a (91-432); 438v1b; 439v1a (91-336); 444i5a; 445i2d1; 459ia; 465v2a.

Xylariaceae II

Colony 2.5 cm diam in 5 days; felty to appressed, azonate; at first white, becoming overlain with a greyish brown mycelium formed by coiled hyphae; margin white, minutely plumose. Occasionally sterile white to white orange tufts forming towards margin of colony; reverse unstained to greyish orange (6B5); no pigment diffusing; remaining sterile.

Total number of isolates: 71.

COMMENTS: The surface of the colony of some isolates of this taxon was radially furrowed; this was reflected in the colony reverse by radial splitting of the agar. A few isolates had yellowish exudate on surface of the colony; other isolates showed white mycelium growing underneath the agar between the lower surface of the agar and the bottom of the Petri dish of the old cultures.

ISOLATES EXAMINED: **Jan 89**: 1v1d; 1v4e; 1v5d; 1i3a; 30i2c; 33v1a (90-20); 37v2c (90-18); 41v1a; 41v1d; 41i1b; 57v1c; 60ic; 69v1e; 86v3c2; 88v2a; 88v2d; 90v2b; 98v1b2; 151v1a; 151v1b; 156v2b (90-22); 186i1c; 198v2c; 216v1e1.

Sep 89: 92v6e (90-187); 94v1b; 98v1a; 98v3c; 103v1c; 108v2d; 109v2c (90-179); 112v1a; 112v2a (90-170); 112v2b (90-163); 112v2c; 112v2e (90-164); 114v3a (90-111); 119i1b; 119v1a; 121v3c (90-165); 123v3b; 130v3a.

Jan 90: 288v2a1; 291v5d; 308v1a (90-226); 308v2a (91-290); 309v1a; 310v1e; 312v1a; 314v2c (90-195); 322v2d; 324i3b; 325v3a (90-209); 331i1a (91-251); 332v2a; 334v1a; 334v2c (91-209) 335v3b (91-155); 336v1b (91-31); 340i3a; 343i2c; 346v6c (91-128); 347i5a.

Sep 90: 363vb; 426v3c; 435v2c (91-534); 436v4e; 436v6a2; 444v3b (91-359); 456i2c; 479vb.

Xylariaceae III

Colony 3 cm diam in 5 days; cottony to felty, faintly zonate; at first white, then pale orange (6A3), later becoming overlain with greyish brown mycelium formed by stromatic hyphae, beginning at the center of colony and progressing towards margin of colony; margin white, scalloped; reverse brownish orange to light brown (6C8 to 6D8); brownish yellow (5C8) pigment diffusing. Neither stromata nor conidiogenous cells produced.

Total number of isolates: 94.

COMMENTS: In old cultures of a few isolates white mycelium grew underneath the bottom of the agar.

According to L. Petrini (pers. comm., 1991) the colony produced by this endophyte matches *Hypoxylon bipapillatum* Berk. & Curt. However, because conidia were not produced by my isolates, I have not assigned them to *H. bipapillatum*.

ISOLATES EXAMINED: **Jan 89**: 1v2a; 4va; 8vc; 18v1a; 27i1a; 37v1a; 39i1a1; 41i2b; 41v1e; 57v4d; 60v1d; 84v2a; 86v2d; 98v2c; 149i1b; 174i1c; 186i1b.

SEP 89: 47v5b; 55i1b; 73v1a; 73v2b; 83v2a; 84v1b; 91i3b; 92v6d; 92i1c2; 92i1d2; 95v2b; 98v1b; 98v3b2 (90-76); 98i5a; 100v2b; 103v2d; 104v5b; 104v6a; 109v1a; 120v1a; 124v2a; 131v1a; 131i1a.

Jan 90: 279v2d; 293v1e; 293v2d; 294i1a; 295v1c; 295v1d; 298i2a; 303i1c (90-227); 307v1a; 307v2a; 309i2c; 312v2a; 314v2d; 317v1a; 317v1b; 319i1d; 323i1a; 323i1b; 323v2d; 325v1e (90-228); 325v2d; 329v3b; 332i3b; 332i6b (91-9); 340v2c; 346v1d; 346v3c; 347i6a.

Sep 90: 362vb (91-537); 363ia; 364v1c; 415i1a; 415v4a; 418i6b; 418v5a; 421i1a; 421v1b; 424v6a; 427va; 428v3a; 433i1a; 435v2b; 435v4d; 435v6d; 436v2c; 436v4d; 439i2a; 441i4a; 441v2c1; 444v5a2; 445v6d (91-538); 453v2d; 472va; 478i1a.

XYLARIACEAE IV

Colony 3.5 cm diam in 5 days; appressed, faintly zonate; at first white, becoming Persian orange (6A7), finally overlain with stone grey (3E2) coiled hyphae forming abundantly in the aerial mycelium at the margin; margin narrowly zonate; occasionally white, fluffy aerial mycelium produced around edge of colony; reverse non pigmented to pale orange (5A3); no pigment diffusing. Neither stromata nor conidiogenous cells produced.

Total number of isolates: 55.

COMMENTS: The colony morphology presented by this endophyte has been found in several members of the Xylariaceae that produce sterile colonies, providing little taxonomically useful information for identification even at the generic level (B. Callan, pers. comm., 1989).

ISOLATES EXAMINED: **Jan 89:** 69v3e; 71v4e1; 75v3c; 86v1b; 90v2c1; 90v3c; 96v2c; 100v2a; 100v2b; 100v2c; 110v1a; 110v1b; 110v2c; 112v1d; 112v1c1; 149v1b; 149v1c; 174v1c1; 216i2c; 221v2c; 223i2a1; 230i2a; 230i2b; 230i2c.

Sep 89: 49v2d; 51i1a; 58v1b; 92v6a; 101v3a; 103v1b; 103v5c; 104i4a; 104i4b; 115i2b; 115i2c; 119v3c; 120v3a; 121v1b.

Jan 90: 244v1a; 308v1b (91-201); 313v2b; 323i3a; 330i1a; 340v3c; 347v4a; 347v4b; 347v4d (91-232).

Sep 90: 428v3b; 436v5c (91-296); 438v1e (91-297); 438v4b; 453v1d2; 467v2d; 480v2e.

XYLARIACEAE V

Colony 3.5 cm diam in 5 days; cottony to velvety, faintly zonate; at first white, to orange white (6A2), eventually overlain with a layer of brownish grey (6F8) mycelium formed of thick stromatic hyphae; margin minutely plumose; reverse dark brown (6B6, 6E3, 6E4); light brown (6D8) pigment diffusing. Neither stromata nor conidiogenous cells observed.

Total number of isolates: 48.

COMMENTS: Stromatic hyphae the same as produced by *X. arbuscula*.

ISOLATES EXAMINED: **Jan 89**: 1v1b; 1v2b; 1v5c; 27v1a; 88v3a; 88i2a; 90v3a; 96v1c; 110v1d; 140i2b; 190v2b; 192v2d; 209i2b; 209i2d; 216i1b.

Sep 89: 3i3a; 91v3a; 93i4a; 97v4d; 98v3a; 100i2a; 100v3a; 100v3d; 103v4b; 104v4a; 109i1a; 118v1a.

Jan 90: 262v2b; 289i2b; 289v2a (91-254); 289v2b; 294v2a1; 295i1a; 297v2e; 302i2a; 302v2a; 303v1b; 304i2b (90-229); 313v2b (91-237); 325v2a; 328v2c (91-148); 334v5c; 335v2a (91-273); 335v3a; 341v3a; 346v2c (91-185).

Sep 90: 417v5b; 478v2a; 479va.

5.2. BASIDIOMYCOTINA

Arthroconidial

OA: Colony 4.5 cm diam in 7 days; cottony, becoming farinaceous in the center, azonate; white to yellowish white (2A2), with colorless to yellowish exudate in the aerial mycelium; margin entire; reverse butter yellow (4A5) to pale orange (5A3). **PYE**: Colony 2 cm diam in 7 days; wooly, slightly raised, azonate; white, with yellowish exudate in the aerial mycelium; margin slightly lobed; reverse butter yellow (4A5). All hyphae staining in phloxine; fertile hyphae frequently septate, branched, to 3.6 µm wide; aerial and submerged hyphae forming hyaline crystals. Arthroconidia formed by septation of undifferentiated lateral branches, cylindrical, one-celled, (4.5-)6.3-8.1(-11) x (-2) 2.5-3.5 µm, hyaline; resistant to Benomyl at 2 ppm. Chlamydospores produced in some isolates, intercalary or terminal, slightly thick-walled, hyaline.

Total number of isolates: 45.

Representative isolates at ATCC: 89-76, 89-104, 91-531, 91-532.

Representative isolates at UAMH (University of Alberta Mold Herbarium and Culture Collection, Edmonton, Alberta): 6671 (KFR 89-73), 6672 (KFR 89-78).

COMMENTS: Although this endophyte was isolated from trees and seedlings located at both subsites, it was more frequent in trees at low várzea. It was recovered from both tissues of the three leaf-age classes.

No major differences were noticed in either medium used except for slower growth rate on PYE.

This endophyte is an arthroconidial anamorph of a basidiomycete and closely resembles the form-genus *Mauginiella* (Group 1, page 370, in Sigler and Carmichael 1976). The isolates were examined by Dr. Lynne Sigler, who agreed with the closeness to *Mauginiella* as was described in Sigler and Carmichael. However, she no longer recognizes this form-genus and there is, as yet, no anamorph name available for these and similar basidiomycetous monokaryons.

ISOLATES EXAMINED: **Jan 89**: 33v2c; 88i2a (89-76); 96i1a; 107i2a (89-79); 120i1a (89-78); 138v2b; 158i2a; 160v2a (89-75); 167v2a (89-77); 184i2a; 194v2a; 194v2b; 209i1b (89-161); 211i2a; 211i2b.

Sep 89: 21i1a; 21i1d; 31v1a; 31v2b; 31v2d (89-104); 35i2a (89-103); 35i2c; 35i2d (90-35); 36i1a; 42v1b; 66v1e; 95i2a (89-105); 95i2c (89-102); 119i3a; 127i5a; 127i5b; 131i3c.

Jan 90: 252i1a (91-111); 259v1b; 262v1b; 265i1a; 267i1a (91-134); 269i1c (90-199); 330i3a.

Sep 90: 351v2a; 351v2b (91-532); 364i1c (91-360); 397i1a; 410i2b; 480v2c (91-531).

5.3. DEUTEROMYCOTINA

5.3.1. COELOMYCETES

HOLOBLASTOMYCETIDAE

Order Blastales

Blastostromatineae

Lasiodiplodia theobromae (Pat.) Griff. & Maubl., Bull. Soc. Mycol. Fr. 25:

57. 1909.

CMD: Colony 5 cm diam. in 5 days; translucent; aerial mycelium scant, azonate; olive brown (4E5); margin even; reverse blackish. Conidiomata formed at margin of colony; stromatic, superficial, globose, dark olivaceous, pubescent, 2-4 mm diam, ostiolate. Conidiophores absent. Conidiogenous cells holoblastic, determinate, discrete, cylindrical, hyaline. Conidia terminal, solitary, subovoid to ellipsoidal, first one-celled, hyaline, thick walled, granulose; mature conidia (18-)24.3-30.3(-34) x 13.2-15.4 µm, two-celled, septum median, with longitudinal striations, dark brown. Teleomorph not produced.

Total number of isolates: 8.

Representative isolates at ATCC: 89-36, 89-163, 91-72, 91-485.

COMMENTS: This endophyte was isolated in higher numbers from seedlings (only one isolate from a tree) located at both subsites (low and high várzea). It was recovered only from the midrib tissues of newly expanded and mature leaves.

This fungus is an important and common tropical plant pathogen. Descriptions and illustrations have been published by Matsushima (1971; 1975), Punithalingam (1976), and Sutton (1980).

RECORDED AS ENDOPHYTE: *Polypodium ciliatum* Willd. (Pteridophyta), *Epidendrum* sp., *Maxillaria* sp., Orchidaceae indet. (Dreyfuss and Petrini 1984).

OTHER SUBSTRATA: plurivorous (including several genera of the Palmae).

DISTRIBUTION: subtropical and tropical.

ISOLATES EXAMINED: **Jan 89**: 90v2a (89-36); 186v1a2.

Sep 89: 78v2d; 101v2a1; 101v4a; 101v5a; 101v5c.

Jan 90: 335v6d.

Sep 90: none.

Pestalotiopsis palmarum (Cooke) Steyaert, Bull. Jard. Bot. Etat Brux. 19(3): 322. 1949.

CMD: Colony reaching 4.5 cm diam in 5 days; translucent; aerial mycelium scant, white, azonate; margin even. Conidiomata scattered, brown, to 220 µm diam, producing a blackish mass of conidia. Conidiophores, hyaline, branched, to 10 long x 4.5 µm. Conidiogenous cells holoblastic, annelidic, indeterminate, integrated, cylindrical, hyaline, with percurrent proliferations. Conidia fusiform, straight, (19.8-)25.2-31.5(-34.2) x 5.4-6.3 µ m, 5-celled, dark brown except for hyaline apical and basal cells; 3-median cells 13.5-16.2 µm long; 2-3 apical appendages to 30 µm long, unbranched; one basal appendage to 5.4 µm long, straight, unbranched.

Total number of isolates: 23.

Representative isolates at ATCC: 89-57, 89-93, 90-52.

COMMENTS: This endophyte was isolated from trees and seedlings, but was more common in seedlings, at both subsites (low and high várzea). It was recovered from the midrib and intervein regions, but was more often found in the midribs of the 3 leaf age classes.

This endophyte matches descriptions of *P. palmarum* reported by Mordue and Holliday (1971) and Simone (1991), and the illustration in Mordue and Holliday (1971).

RECORDED AS ENDOPHYTE: in *L. ramsayi* (Rodrigues and Samuels 1990).

OTHER SUBSTRATA: on Palmae: *Areca, Arenga, Astrocaryum, Borassus, Caryota, Chamaerops, Chrysalidocarpus, Cocos, Elaeis, Howea, Pandanus, Phoenix, Rhapis, Roystonia, Sabal, Washingtonia.* On *Capsicum, Hevea, Manilkara, Musa.*

DISTRIBUTION: subtropical and tropical.

ISOLATES EXAMINED: **Jan 89**: 10va; 49i3b; 60v2a (89-57); 80v3a; 84v3a (89-3); 156v2a; 156v2e1; 156v2e2; 186v1c; 216v2a.

Sep 89: 13v1c; 100i1a; 100v1c; 100v1d; 100v5b; 101i2a; 101i3a; 101v2a2 (90-69); 101v2b; 101v6a (90-90); 101v6b; 128i1c (90-52); 132i3a (90-91).

Jan 90: none. **Sep 90**: none.

ENTEROBLASTOMYCETIDAE

Order Phialidales

Phialopycnidiineae

Phoma sp.

CMD: Colony 1.5 cm diam in 5 days; translucent to opaque, faintly zonate; woolly, whitish first, then darkening to greenish brown in the center of colony; margin white, even. Conidiomata pycnidial, produced in center of colony, immersed to semi-immersed, dark brown, globose to sub-globose, 189-308 μm wide, ostiolate. Conidiophores absent. Conidiogenous cells enteroblastic, phialidic, discrete, ampulliform, hyaline, 4-5 x 2-2.5 μm. Conidia oblong, subglobose to ellipsoidal, 2.7-3.6(-4.5) x 1.8-2.7 μm, one-celled, eventually 2-celled with septum median, hyaline to subhyaline, indistinctly guttulate.

Total number of isolates: 37.

Representative isolates at ATCC: 89-129, 91-44, 91-292, 91-474.

COMMENTS: This endophyte was isolated from trees and seedlings at both low and high várzea but with greater frequency in trees and seedlings located at low várzea. It was recovered from midrib and intervein regions of the three leaf age classes.

ISOLATES EXAMINED: **Jan 89**: 37i2b; 47v1c; 53i1a; 53i1b; 53i1c; 66i2a; 66v2d; 69v1a; 69v1c; 69v1d; 73v1b; 75i2d2; 75v1b; 149i1d; 158v2c (89-41); 216v2e2; 228i1a (90-50; 90-80).

Sep 89: 13v1b (91-44); 17v1a (91-42); 23v1c (91-43); 59v2e (91-17); 62v2a; 83i2a (91-566); 90v2b; 102v1a (91-46).

Jan 90: 243v2d; 264v2c; 267i1b; 287i4a (91-292); 315i3a (91-492); 333v3a (91-489).

Sep 90: 366va (91-474); 403v1a; 417v5d (91-473); 432v4a1; 445v3d1; 451i2e.

Phialostromatineae

Colletotrichum gloeosporioides (Penz.) Sacc., Fung. Agrum. 2: 6. 1882.

CMD: Colony 4 cm diam in 5 days; translucent to opaque, azonate; aerial mycelium white, occasionally darkening to pale brown in the center; margin even; reverse non pigmented. Conidiomata superficial, brown, pubescent, 198-264 x 198-264 µm, producing a pale orange to salmon mass of conidia. Conidiophores subhyaline, branched and septate. Conidiogenous cells enteroblastic, phialidic, hyaline, determinate, cylindrical, integrated, to 31 µm long. Setae abundant, brown, smooth, septate, 59.4-90 x 2.7-3.6 µm. Conidia cylindrical, (10.8-)13.8-19.3(-24.5) x (3.6-)4.5-6.3(-7) µm, one-celled, hyaline, minutely guttulate. Teleomorph not produced.

Total number of isolates: 58.

Representative isolates at ATCC: 89-56, 91-287, 91-482.

COMMENTS: This endophyte was isolated from trees and seedlings located at high and low várzea. It was recovered from both tissues but was more often found in the midrib region of the 3 leaf age classes.

This endophyte agrees with descriptions and illustrations of *C. gloeosporioides* given by Arx (1957) and Mordue (1971).

RECORDED AS ENDOPHYTE: in *Arctostaphylos uva-ursi* (Widler and Müller 1984); *Suaeda fruticosa* Forskal (Fisher and Petrini 1987a); *Alnus glutinosa* (Fisher and Petrini 1990); *Anthurium* sp, *Philodendron* sp, *Epidendrum* sp, Araceae indet. (Dreyfuss and Petrini 1984).

OTHER SUBSTRATA: plurivorous.

DISTRIBUTION: cosmopolitan.

ISOLATES EXAMINED: **Jan 89**: 4vb; 4vc; 30v2c; 37i2a; 47v1b; 57i4a; 60v3b; 63v1a; 63v2a; 63v3a; 63v3c; 75v1c; 84v2b; 86v1a; 86v3b; 138v3a; 149v2b; 174i1d; 174v2a; 184v1a; 186i1a1; 186i1d; 200i2a; 209v2a; 216v1b1; 218v2b2; 221v1a; 221v1c; 221v1d.

Sep 89: 7v2d; 74v1b; 76v1c; 89i2a; 92v2d; 94i1c; 94i1d; 94i3e; 94v6e; 100v5a; 101i6b; 106v3c; 107v1c; 107v1d; 107v2a; 107v2b; 108i1d.

Jan 90: 241v1c; 263v2a; 263v2c;264v2e1; 265v1b; 291v1e; 297i2b1; 298v1a.

Sep 90: 432v5b; 438i2b; 445i5e; 478i2d.

Colletotrichum sp

CMD: Colony 3 cm diam in 5 days; opaque, azonate; greenish brown with aerial mycelium pale brown; margin even; reverse blackish. Conidiomata superficial, dark brown, 712-801 x 623-712 µm. Conidiophores pale brown, branched and septate, to 81 µm long x 7.2 µm wide. Conidiogenous cells enteroblastic, phialidic, subhyaline, determinate, cylindrical, integrated. Setae brown, smooth, septate, 74.7-90 µm long x 2.7 µm wide. Conidia cylindrical, (19.8-) 21.3-26.8 (-31.9) x (5.4-) 6.5-10.6 (-11.8) µm, one-celled, sub-hyaline, minutely guttulate. Teleomorph not produced.

Total number of isolates: 10.

Representative isolate at ATCC: 91-510.

COMMENTS: This endophyte was isolated from trees and seedlings located at low and high várzea. It was recovered from midrib and intervein regions of newly expanded and mature leaves.

ISOLATES EXAMINED: **Jan 89**: none. **Sep 89**: none.

Jan 90: 264v2e2; 284v1a (91-347); 288v2d.

Sep 90: 394vb; 432i4b; 432i4c; 432v4a2; 438i4a; 438v5c (91-510); 439v2b.

Phomopsis sp. 1

CZ: Colony 4 cm diam in 5 days; opaque, at first white, then darkening to orange white (6A2) to flesh (6B3) in the center, white elsewhere; margin finely plumose; reverse unstained to rosewood (9D5). Conidiomata eustromatic, abundant, semi-immersed, dark brown, producing a yellowish mass of conidia. Ostiole single, occasionally papillate. Conidiophores hyaline, branched and septate. Conidiogenous cells enteroblastic, phialidic, integrated, determinate, cylindrical, hyaline. Conidia fusiform, (4.5-)5.4-6.3 x 1.8-2.7 µm, one-celled, hyaline, biguttulate, smooth. β-conidia hamate, one-celled, hyaline, eguttulate. Teleomorph not produced.

Total number of isolates: 97.

Representative isolates at ATCC: 91-478, 91-547.

COMMENTS: This endophyte was isolated from trees and seedlings at low and high várzea, but was more frequently found in the high subsite. It was most commonly recovered from the midrib region of the 3 leaf age classes.

This taxon resembles *Phomopsis* sp. 3 described as an endophyte of the palm *Licuala ramsayi* by Rodrigues and Samuels (1990).

ISOLATES EXAMINED: **Jan 89**: 30v2a; 63v2b; 75v2e2; 83v3d2; 86v3c1; 110v1e; 110v2a; 110v2b; 112v1a; 112v1e; 169v1a; 169v1c; 169v2a; 169v2b; 171v2a; 171v2b; 174i1a; 176v1d; 190v2c; 216i1a; 216i2b; 216v1b2; 216v1d; 216v2b; 216v2e1; 223v1a; 223v1b; 225v1a; 225v2a; 225v2b; 230v1b (90-140); 232v2a; 232v2b; 234v1b; 234v1d.

Sep 89: 23v1a; 47v1a; 51v2c; 58v2d (91-76); 59v2d; 83v1a; 83v2b; 85v2a; 87v2d; 92v2e; 92v3e; 92v5b2; 94v3b (91-45); 94v6c; 100i3b; 100i6a; 111v1a.

Jan 90: 243v2c; 263v2d; 264v2a; 264v2b; 273v1a; 277v1a; 278v2a; 281v2e; 291v3e (91-214); 291v4a (91-217); 294i2b (91-270); 298i2c (91-490); 299v5a (91-215); 299v6b; 332v5e (91-269); 342v6a.

Sep 90: 364v1d1; 365v1b; 367vb (91-556); 369va; 370va (91-439); 371v1a (91-476); 371v1b (91-475); 375i1a (91-547); 386v2a; 387i1a (91-480); 387v2b; 404v2c; 404v2d; 405va; 413v2a; 417v3a1; 418i3b; 432i1c; 435i2b (91-438); 435i2c; 438v5b; 442i1c (91-477); 444i6d2; 447v2e (91-478); 459va (91-479); 460v1a; 461va; 467v2c; 483v2b.

Phomopsis sp. 2

CZ: Colony 4 cm diam in 5 days; opaque, mycelium appressed, champagne (4B4); reverse brownish yellow (5C8). Conidiomata eustromatic, scattered, immersed, dark brown, less then 1 mm diam., producing a pale grey mass of conidia. Ostiole single. Conidiophores hyaline, branched and septate. Conidiogenous cells enteroblastic, phialidic, integrated, determinate, cylindrical, hyaline. Conidia, fusiform, 6.3-7.2(-8.1) x 2.7-3.6 µm, one-celled, hyaline, biguttulate, smooth. β-conidia hamate, one-celled, hyaline, eguttulate. Teleomorph not produced.

Total number of isolates: 7.

Representative isolates at ATCC: 89-10, 89-11.

COMMENTS: An endophyte isolated from trees and seedlings at low and high várzea, found only in the midrib region of the 3 leaf age classes.

It resembles *Phomopsis* sp.1 described from *Licuala ramsayi* (Rodrigues and Samuels 1990).

ISOLATES EXAMINED: **Jan 89**: 60v1a (90-139); 86v1c (89-11); 86v1d; 107v2d (89-10); 107v2e; 176v1a.

Sep 89: none.

Jan 90: 299v6a (91-216).

Sep 90: none.

5.3.2. HYPHOMYCETES

Acremonium sp

CMD: Colony 1.5 cm diam in 5 days; translucent, narrowly zonate; aerial mycelium scant, white; margin even; colorless droplets forming abundantly in the aerial mycelium. Conidiogenous cells enteroblastic, phialidic, hyaline, smooth, cylindrical, basally septate, simple or occasionally branching, to 37 µm long x 1.8-2.7 µm wide; collarette indistinct. Conidia cylindrical with rounded ends, (3.6-)4.1-5.5(-6.3) x 1.4-2.3(-2.7) µm, one-celled, hyaline, smooth. Teleomorph not produced.

Total number of isolates: 10.

Representative isolates at ATCC: 89-23, 89-159.

COMMENTS: This endophyte was isolated mainly from trees at low várzea, except for one isolate at high várzea. It occurred mainly in the midrib tissue, except for one isolate that was recovered from the intervein region, of all three leaf age classes.

ISOLATES EXAMINED: **Jan 89**: 14v3a (89-22); 21v1a; 21v2a; 21v2b; 47i2a; 84v1c (89-71); 167v1a (89-23).

Sep 89: 10v1a; 75v1a.

Jan 90: 328v3c.

Sep 90: none.

Acrodictys elaeidicola M. B. Ellis, Mycol. Pap. 79: 7. 1961.

(Plate XXIII, Figs. 1-2)

CMD: Colony reaching a maximum of 1.5 cm diam in 5 days; opaque, azonate; pale brown to dark brown; margin white, even. Conidiophores macronematous, mononematous, straight or flexuous, brown, smooth, septate, 58.5-90 (-135) µm long x 3.6-5.4 µm. Conidiogenous cells monoblastic, integrated, terminal, proliferating percurrently. Conidia pyriform, (17.6-) 19.7-25.9 (-31.9) x (10.8-) 11.2-13.1 (-13.5) µm, dictyosporous with 2-3 transverse septa and 1-longitudinal septum, brown, except for pale basal cell, smooth, solitary, terminal.

Total number of isolates: 26.

Representative isolates at ATCC: 91-568, 91-569, 91-575, 91-576.

COMMENTS: *Acrodictys elaeidicola* was present both in trees and seedlings, but it was most often recovered from seedlings at low várzea. It was present in the midrib as well as in the intervein regions both of newly expanded and mature leaves.

This fungus matches the description and illustrations of *A. elaeidicola* (Ellis 1961), a species originally described from a species of a genus of palm, *Elaeis*.

RECORDED AS ENDOPHYTE: none.

DISTRIBUTION: Ghana, Sierra Leone.

ISOLATES EXAMINED: **Jan 89:** 37v1c; 81v3a; 107i1a; 149v1e; 151i2b; 218i1a.

Sep 89: 115v1b; 115v3d (91-97); 124i3b (91-23); 128v1b.

Jan 90: 265v2b.

Sep 90: 415v6a (91-575); 417i4b (91-591); 420v3c (91-576); 423i5e; 423v6a; 424i2a; 424i3a; 424v4e; 426v2a1 (91-569); 426v3b1; 426v3d1; 427v1c; 428v2c (91-574); 429v3b (91-573); 429v3c.

Chloridium* ?*preussii W. Gams & Hol.-Jech., Stud. Mycol. 13: 35. 1976.

(Plate XXIV, Figs. 1-3)

CMD: Colony reaching maximum of 1 cm diam in 5 days; opaque, woolly, faintly zonate; olive brown; margin broadly lobed. Conidiophores macronematous, mononematous, erect, unbranched, pale brown, smooth, to 50 µm long x 2.7-3.6 µm wide. Conidiogenous cells enteroblastic, phialidic, integrated, terminal, proliferating percurrently. Conidia formed in slimy masses, subglobose, (1.8-)2.3-3.2(-4) x (1.4-)1.8-2.5(-3) µm, one-celled, hyaline, smooth. Chlamydospores in chains, thick walled, pale brown. Teleomorph not produced.

Total number of isolates: 4.

Representative isolates at ATCC: 90-79, 91-493, 91-494.

COMMENTS: *Chloridium* ? *preussii* was isolated exclusively from trees at high várzea. It was equally found in the midrib and intervein regions of newly expanded and mature leaves.

This differs from *Chloridium preussii* as described by Gams and Holubová-Jechová (1976), which has ellipsoidal conidia and lacks chlamydospores.

RECORDED AS ENDOPHYTE: none.

OTHER SUBSTRATA: *Acer*, *Alnus*, *Fraxinus*, *Quercus*.

DISTRIBUTION: temperate.

ISOLATES EXAMINED: **Jan 89**: 225v1b (90-79).

Sep 89: none.

Jan 90: 280v1d; 288i2b.

Sep 90: 449i2d (91-592).

Curvularia pallescens Boedijn, Bull. Jard. Bot. Buitenzorg Ser. 3, 13: 127. 1933.

CMD: Colony 3.5 cm diam in 5 days; opaque, velvety, faintly zonate; dark brown; margin entire. Conidiophores macronematous, mononematous, branched, straight to flexuous, slightly geniculate, pale brown, smooth, > 90 μm long. Conidiogenous cells enteroblastic, polytretic, integrated, terminal. Conidia ellipsoidal, (21-) 25.5-32(-33) x (8.4-)10.1-12.8(-14.3) μm, 4-celled, pale brown, straight to slightly curved, smooth.

Total number of isolates: 10.

COMMENTS: This endophyte was isolated from trees and seedlings at both subsites, but was found more often in trees and at low várzea. It was recovered most frequently from the intervein region of the three leaf age classes.

The palm isolates agree with descriptions and illustrations of *C. pallescens* given by Ellis (1966). *Curvularia pallescens* is a ubiquitous fungus with a cosmopolitan distribution, being found on many unrelated plant families, including Palmae.

RECORDED AS ENDOPHYTE: in Araceae (Petrini and Dreyfuss 1981).

OTHER SUBSTRATA: on *Agave*, *Arachis*, *Axonopus*, *Borassus*, *Brachiaria*, *Brassica*, *Calotropis*, *Cassia*, *Centrosema*, *Coelorrhachis*, *Coix*, *Cynodon*, *Digitaria*, *Echinochloa*, *Elaeis*, *Euchlaena*, *Musa*, *Oryza*,*Pennisetum*, *Physalis*, *Saccharum*, *Sorghum*, *Sporobolus*, *Triticum*, *Zea*, *Zingiber*.

DISTRIBUTION: tropical.

ISOLATES EXAMINED: **Jan 89**: 55i1a (89-04); 55v2a; 81i3a; 192i1a.
Sep 89: 2v2a; 7i2a; 10i3a; 68v1c (90-92); 97i1b.
Jan 90: 241i1b (91-263).
Sep 90: none.

Fusarium oxysporum Schlecht., Flora Berl. 2: 139. 1824.

PDA: Colony 8 cm diam. in 5 days; floccose to felty, faintly zonate; first white then pale orange; margin fringed; pigment diffusing pale purple. **SNA**: Conidiophores forming in the aerial mycelium, branched, hyaline. Conidiogenous cells phialidic, cylindrical, hyaline. Microconidia formed on monophialides in the aerial mycelium, ellipsoidal, one-celled, hyaline. Macroconidia in the aerial mycelium, fusiform, with a foot cell, 3-5 septate, hyaline.

Microconidia: 5.4-12 x 2.7-3.6 µm,
3-septate: 30.6-36-45 x 3.6-4.5 µm,
4-septate: 43.2-46.8 x 3.6-4.5 µm,
5-septate: 45-58.5 x 3.6-5.4 µm.

Total number of isolates: 6.

COMMENTS: This endophyte was isolated mainly from trees, with only 1 isolate recovered from a seedling, exclusively at low várzea. It was recovered only from midrib tissue of newly expanded and mature leaves.

My isolates agree with the descriptions and illustrations of *F. oxysporum* given by Booth (1971) and Nelson et al. (1983).

RECORDED AS ENDOPHYTE: in Araceae, Bromeliaceae, Orchidaceae (Petrini and Dreyfuss 1981); *Epidendron porpax* Reichb., *Peperomia* cf. *reflexa* A. Dietr. (Dreyfuss and Petrini 1984); *Oryza sativa* L. (Fisher and Petrini 1992).

OTHER SUBSTRATA: plurivorous.

DISTRIBUTION: cosmopolitan.

Jan 89: none.

Sep 89: 13v2a; 39v1a.

Jan 90: 335v5d.

Sep 90: 357va (91-392); 378v1b; 378v1c.

Fusarium sacchari (Butler) W.Gams var. ***elongatum*** Nirenberg, Mitt. Biol. Bundesanst. Land-Forstwirtsch. Berlin-Dahlem 169: 59. 1976.

PDA: Colony 6.5 cm diam in 5 days; cottony, azonate; cream (4A3); margin even. **SNA**: Conidiophores in the aerial mycelium, branched, hyaline. Conidiogenous cells monophialidic, occasionaly polyphialidic, cylindrical, hyaline, 22.5-31.5 µm long x 2.2-2.7 µm wide. Microconidia formed in false heads, oval, one-celled, hyaline. Macroconidia straight to falcate, with distinct foot cell, 3-5 septate, hyaline.

Microconidia: 9.9-11-12.1 x 1.8-2.7-3.6 µm,
3 septate: 37.5-48.4-51 x 2.7-3.6 µm,
5 septate: 62.7-67.5-77 x 2.7-3.6-4.5 µm.

Total number of isolates: 6.

Representative isolate at ATCC: 90-47.

COMMENTS: This endophyte was isolated only from a single sampling period (Jan 89). It was found in trees located at low várzea, except for one isolate of a seedling at high várzea, where it occurred both in midrib and intervein regions of the three leaf age classes.

This endophyte agrees with descriptions and illustrations of *F. sacchari* var. *elongatum* given by Gerlach and Nirenberg (1982).

RECORDED AS ENDOPHYTE: none.

OTHER SUBSTRATA: on *Saccharum*, *Zea*.

DISTRIBUTION: subtropical and tropical.

ISOLATES EXAMINED: **Jan 89**: 151i1b; 165v1a (89-38); 165v1b; 165v1c; 165v2a; 174i2d1.

Sep 89: none. **Jan 90**: none. **Sep 90**: none.

Fusarium semitectum Berk. & Rav. in Berkeley var. ***majus*** Wollenw., Z. Parasitenk. 3: 325-327. 1931.

PDA: Colony 7 cm diam in 5 days; floccose, azonate; white to pale salmon; margin even. **SNA**: Conidiophores in the aerial mycelium, branched, hyaline. Conidiogenous cells mostly monophialidic, cylindrical, sympodially proliferating, to 9 µm long x 2.7-3.6 µm. Microconidia uncommon, in the aerial mycelium. Macroconidia produced in the aerial mycelium, fusiforme, 3-5 septate, hyaline.

Microconidia: 6.3-8.1 x 2.7 µm,

3 septate: 25.5-34.5 x 3.6-4.5 µm,
5 septate: 42-52.5 x 4.5-5.4 µm.

Total number of isolates: 3.

Representative isolates at ATCC: 90-49.

COMMENTS: This endophyte was isolated from trees located at low várzea, where it was recovered from the midrib region of unopened and newly expanded leaves.

My isolates agree with descriptions and illustrations of *F. semitectum* var. *majus* given by Gerlach and Nirenberg (1982).

RECORDED AS ENDOPHYTE: none.

OTHER SUBSTRATA: *Beta*, *Elaeis*, *Ipomoea*, *Juglans*, *Musa*.

DISTRIBUTION: subtropical and tropical, occasionally in temperate regions.

ISOLATES EXAMINED: **Jan 89**: 158v1a (89-113).
Sep 89: none.
Jan 90: 243v2a; 243v2b. **Sep 90**: none.

Fusarium verticillioides (Sacc.) Nirenberg, Mitt. Biol. Bundesanst. Land-Forstwirtsch. Berlin-Dahlem 169: 26. 1976.

PDA: Colony reaching 7.5 cm diam in 5 days; cottony, azonate; cream, margin even; reverse vinaceous. **SNA**: Conidiophores in the aerial mycelium, branched, hyaline. Conidiogenous cells monophialidic, almost cylindrical, to 27 µm long x 3.6 µm wide. Microconidia clavate with flat base, one-celled. Macroconidia present in the aerial mycelium, straight to falcate, eventually with foot cell, 3-4-5 septate, hyaline.

Microconidia:	6.3-9 x 1.8-2.7 µm,
3 septate:	39.6-51 x 1.8-2.7 µm,
4 septate:	52.5-64.5 x 2.7-3.6 µm,
5 septate:	68.2-77 x 2.7-3.6 µm.

Total number of isolates: 15.

Representative isolates at ATCC: 89-20.

COMMENTS: *Fusarium verticillioides* was isolated from trees and seedlings at both subsites, however, it was more frequently isolated from the low várzea. Isolates were recovered from midrib and intervein tissues of the three leaf age classes.

This endophyte agrees with descriptions and illustrations of *F. verticillioides* given by Gerlach and Nirenberg (1982).

RECORDED AS ENDOPHYTE: none.

OTHER SUBSTRATA: *Oryza, Saccharum, Zea.*

DISTRIBUTION: subtropical, and tropical.

ISOLATES EXAMINED: **Jan 89**: 13i2d (89-20); 145v1 (89-60); 145v1b; 158v2d.

Sep 89: 13v1a; 31v2a; 94i1a (90-158).

Jan 90: 265v1a; 280v2a; 284i1a; 326i1a; 336v4a; 336v4c.

Sep 90: 427i2b; 427i2c.

Idriella amazonica K. F. Rodrigues & Samuels, Mycotaxon 63: 275. 1992.

PDA: Colony 5 cm diam in 5 days; opaque, aerial mycelium scant, radially furrowed at center, azonate; pale orange then darkening to brown from the center outwards; margin colorless; reverse light brown, agar shallowly spliting in a radial pattern. Conidiophores macronematous, mononematous, 28.8-45 x 2.7-3.6 µm, branched, sub-hyaline to brown, produced on pale orange to dark brown sporodochia. Conidiogenous cells integrated, sympodial, denticulate. Conidia lunate (18-)20-26(-27) x (3.5-) 3.7-4.6(-4.5) µm, 1-4-celled, sub-hyaline to olive brown. Chlamydospores intercalary to terminal, ellipsoidal to subglobose, brown, 6.3-9 µm broad.
Total number of isolates: 6.
Representative isolates at ATCC: 90-154.
COMMENTS: This endophyte was isolated only from seedlings located at low and high várzea. It was recovered from midrib and intervein regions of mature and newly expanded leaves.
ISOLATES EXAMINED: **Jan 89**: 84i1c; 86i1a; 88i2b (90-46).
Sep 89: 94v5b (91-98); 100i4c; 121v3b (90-154).
Jan 90: none. **Sep 90**: none.

Idriella asaicola K. F. Rodrigues & Samuels, Mycotaxon 63: 274. 1992.

PDA: Colony 5.5 cm diam in 5 days; opaque, appressed with scattered white tufts in the center, azonate, pale salmon, then forming brown patches from the center outwards; margin colorless; reverse brown. Conidiophores solitary, to 30 µm long, or branched, hyaline first, then yellowish brown, 27.9-63.9 x 1.8-2.7 µm, produced in tufts. Conidiogenous cells integrated, sympodial, denticulate. Conidia lunate, (11.6-)12-16.3(-20.7) x (2.7-)2.5-3.3(-3.6) µm, 1-2 celled, hyaline. Chlamydospores intercalary to terminal, elliptical to subglobose, brown, guttulate, 4.5-6.3 µm broad.

Total number of isolates: 6.

Representative isolate at ATCC: 90-143.

COMMENTS: This endophyte was isolated solely from trees located at low várzea. It was recovered from the midrib region of unopened and newly expanded leaves.

ISOLATES EXAMINED: **JAN 89**: 160v2b.

Sep 89: 19v2a1 (90-143); 19v2a2.

Jan 90: none.

Sep 90: 402va2; 411v2a1; 411v2a2 (91-553).

Idriella euterpes K. F. Rodrigues & Samuels, Mycotaxon 63: 272. 1992.

PDA: Colony 4.5 cm diam in 5 days; cottony, azonate; pale orange to orange; margin fringed; reverse pale orange, agar radially splitting. Conidiophore branched, hyaline, 18-26.1 x 2.7 µm, produced on orange sporodochia, abundantly formed in the center of colony. Conidiogenous cells integrated, lageniform, hyaline, monoblastic or sympodially proliferating.

Conidia falcate, (38.7-)45.5-63.1(-64.8) x (1.4-)1.6-2.6(-3.3) µm, 1-2-celled, hyaline. Chlamydospores intercalary, sub-globose, sub-hyaline to brownish, 6.6-8.8 µm broad.

Total number of isolates: 61.

Representative isolate at ATCC: 91-285.

COMMENTS: This endophyte was isolated almost equally from trees and seedlings, primarily located at low várzea. It was recovered from both tissues and was present in the three leaf-age classes.

ISOLATES EXAMINED: **JAN 89**: 6ia (89-48); 6ib6 (90-44); 8ia; 33i1c; 33i1d; 33i2a (90-28); 33i2c; 33v1c; 33v1e; 35i1b (89-52); 37i1a (89-49); 37v1b1 (90-33); 39i2a; 41i2c; 41i2d (90-32); 57i1a; 57v2b1 (90-25); 63v3e; 69i1b; 69i2a; 69i2b; 84i2b; 86i2a2; 86i3c (90-26); 86i3d2; 88i3a2 (90-27); 90i3a; 151i1a; 190i2c (89-21); 221i1a; 223v2a; 223v2b.

Sep 89: 39v1b (90-54); 44v2a (90-55); 78v1c (91-16); 92v1c1 (90-184); 108i1b (91-75); 108i1c; 108i3b; 115v1d (90-155); 128i2a; 128v2e; 128v3d (90-176).

Jan 90: 270v1a (91-491); 320v3a; 330v2e (91-240); 336v4a; 346v4e (91-285).

Sep 90: 364i1b; 364i2a (91-483); 364i2b; 364i2c; 364v2d (91-391); 397i1b (91-557); 408ib; 410v2a (91-559); 420i1a; 420i1b; 420i1c; 420i3c; 432v6e (91-558).

Mycoleptodiscus **sp.**

CMD: Colony 8 cm diam in 5 days; aerial mycelium scant, faintly zonate, margin hyaline, even; reverse olive brown. Sporodochia produced at center of colony, superficial, yellowish brown, circular, with conidiogenous cells arranged radially, clypeate, forming a white mass of conidia. Conidiogenous cells enteroblastic, phialidic, determinate, discrete, ampulliform, pale brown, smooth, to 18 µm long x 6.3 µm wide. Conidia hyaline, minutely guttulate, 2-celled, septum median, cylindrical, (33-)35-40(-45) x (7-)8-10(-12) µm, with an unbranched appendage at each end, 21-27.5 µm long.

Total number of isolates: 2.

COMMENTS: *Mycoleptodiscus* sp. was isolated from only one sampling period (Jan 89) from a seedling located at low várzea. It was recovered from the midrib and intervein regions of a mature leaf.

Three species of *Mycoleptodiscus* are known to have 2-celled conidia, viz. *M. brasiliensis* Sutton & Hodges, *M. sphaericus* Ostazeski, and *M. terrestris* (Gerdemann) Ostazeski. *Mycoleptodiscus terrestris* is closest to my isolates because of the presence both of apical and basal appendages on the conidia. However, *M. terrestris* has smaller conidia (20-35 x 4.5-7 µm) and shorter appendages (9-18 µm long) [Sutton 1973; Sutton and Alcorn 1990].

ISOLATES EXAMINED: **Jan 89**: 75i1d; 75v1a.

Sep 89: none. **Jan 90**: none. **Sep 90**: none.

Nigrospora sphaerica (Sacc.) E. Mason, Trans. Br. Mycol. Soc. 12: 158. 1927.

CMD: Colony 2.5 cm diam in 5 days; aerial mycelium scant; margin hyaline. Conidiophores branched, sub-hyaline to pale brown, smooth. Conidiogenous cells monoblastic, determinate, discrete, ampulliform, hyaline, to 10 μm diam. Conidia spherical, 12-18(-20) μm diam, one-celled, black, smooth.

Total number of isolates: 2.

COMMENTS: *Nigrospora sphaerica* was isolated from a single sampling period (Jan 89), from a tree, and a seedling located at low várzea. It was found in the midrib tissue of mature leaves. It is a ubiquitous species found in a wide geographic range, especially in the tropics, on many different host plants.

RECORDED AS ENDOPHYTE: in *Peperomia* sp. (Dreyfuss and Petrini 1984);.*Chamaecyparis thyoides* (Bills and Polishook 1992).

OTHER SUBSTRATA: *Acacia*, *Agropyron*, *Areca*, *Avena*, *Bambusa*, *Bauhinia*, *Beta*, *Carya*, *Cocos*, *Cynodon*, *Eichhornia*, *Elaeis*, *Gossypium*, *Helianthus*, *Juncus*, *Malus*, *Quercus*, *Secale*, *Setaria*, *Sorghum*, *Stenotaphrum*, *Tridens*, *Triticum*, *Zea*.

DISTRIBUTION: cosmopolitan.

ISOLATES EXAMINED: **Jan 89**: 35v2a (89-02); 84v3c. **Sep 89**: none. **Jan 90**: none. **Sep 90**: none.

Thozetella sp. (Plate XXV, Figs. 1-3)

CMD: Colony 1 cm diam in 5 days; opaque, pale brown, scant aerial mycelium, faintly zonate; margin colorless, narrowly lobed; reverse unstained. Conidiomata synnematal, white, formed within 10 days of incubation, to 90 µm high. Conidiophores branched, septate. Conidiogenous cells phialidic, integrated, cylindrical, subhyaline to pale brown, 17.5-24.5 x 3.5 µm. Conidia fusoid to falcate, (19.8-)22.8-27.5(-27.9) x 2.7-3.6 µm, one-celled, with an apical and basal appendage, to 9 µm long, hyaline, minutely guttulate, smooth. Chlamydospores thick walled, spherical to oval, intercalary and terminal, guttulate, pale brown.

Total number of isolates: 125.

Representative isolates at ATCC: 90-174, 91-549, 91-550, 91-567.

COMMENTS: This endophyte was isolated mainly from seedlings located at low várzea; occurring almost equally in midrib and intervein tissues of newly expanded and mature leaves. One isolate was recovered from an unopened leaf.

ISOLATES EXAMINED: **Jan 89**: 30v2e; 57i4c; 63i2b (89-63); 66i2e; 66v2b; 72v3e2 (89-64); 75v3b; 75v3d; 86i1d1; 88i2c; 88i2d1; 90i1d (89-40); 96v2b; 96v2d; 98v2b; 100i2a; 103i1c; 151i2a; 200i1b; 221v2a.

Sep 89: 91i1b; 92i5d; 92i5e; 92i6d; 94i4d; 95i5b; 95v5b; 98i3b; 109v3b; 112i1a; 112i1b; 112i3d1 (90-188); 113i1a; 113i1b; 113i2b; 113i3a2; 113i3b2; 113i3d; 113v1c; 114i1a; 114v1c; 115v2b (90-160); 115v2c; 118i3a; 119i2a; 120i1a; 120i3d; 124i2a; 124i3e (90-89); 124v2c; 128i2b;

128i2c; 128v2d; 129v1a; 129v1b; 129v2d; 129v3b; 129v3c; 129v3e; 130v1a; 131i2a; 131v1b; 131v1d.

Jan 90: 291v6a1; 314i1b; 315i1d; 315i2d; 318i2e; 318i3c; 320i2b; 327i3c; 327v1a; 327v1b; 327v1c; 327v2e; 328v2d; 329v2e; 329v3d; 330i2b; 330v3c; 330v3d; 331v2a; 331v2b; 346i1c; 347i2a.

Sep 90: 414i3a; 414i5a2; 414v2b; 414v2c; 423i1a; 423i1b; 423i4b; 423i5a; 423i6a; 423v1a; 423v1b; 423v1c; 423v1d; 423v2a (91-551); 423v3d; 423v3e; 423v5d; 423v5e; 423v6d; 423v6e; 424i2c; 424i3d; 424i6a; 424i6b (91-549); 424i6c; 424v1a; 424v1b; 424v1c; 424v2a1; 424v3b; 424v5b; 427i3c; 432i3a (91-495); 432i4a; 432i5a; 432i5b; 433v3c; 435i3a; 435i4a; 435i4b; 435v6a; 469v2a; 483v1a.

Wardomyces sp. (Plate XXVI, Figs. 1-2)

CMD: Colony reaching 2.5 cm diam in 7 days; translucent to opaque, azonate; aerial mycelium scant, greenish brown; margin colorless. Conidiophores semi-macronematous, mononematous, hyaline, occasionally branching, 6.3-13.3 μm long x 2.8-3.5 μm. Conidiogenous cells polyblastic, integrated, determinate, ampulliform, hyaline. Conidia solitary, ovoid to ellipsoidal, (5.4-)5.8-9.1(-9.9) x (2.7-)3.5-5.4(-6.3) μm, with a longitudinal germ slit, one-celled, dark brown, smooth.

Total number of isolates: 12.

Representative isolates at ATCC: 89-66, 91-12, 91-13.

COMMENTS: This endophyte was isolated from trees and seedlings at low and high várzea. It was found in midrib and intervein tissues but showed a higher frequency in the intervein region of the three leaf age classes.

Most described *Wardomyces* species have penicillate conidiophores bearing clusters of conidia at the apex. The conidiophores of my isolates were short and inflated. Despite this difference, the black conidia with a longitudinal germ slit indicate that this is a species of *Wardomyces*.

ISOLATES EXAMINED: **Jan 89**: 44v3b (89-55); 47v1a (89-66); 63v2d.

Sep 89: 64i2a (90-181); 97i6d; 99i5a (90-183); 100i1c; 107i2d; 109i3e; 116i2a; 129i2c.

Jan 90: 287i6a.

Sep 90: none.

PLATE I

Figs. 1-2. *Letendraea* sp.

Fig. 1. Ascospores.

Fig. 2. Ascus.

Scale bar = 10 µm

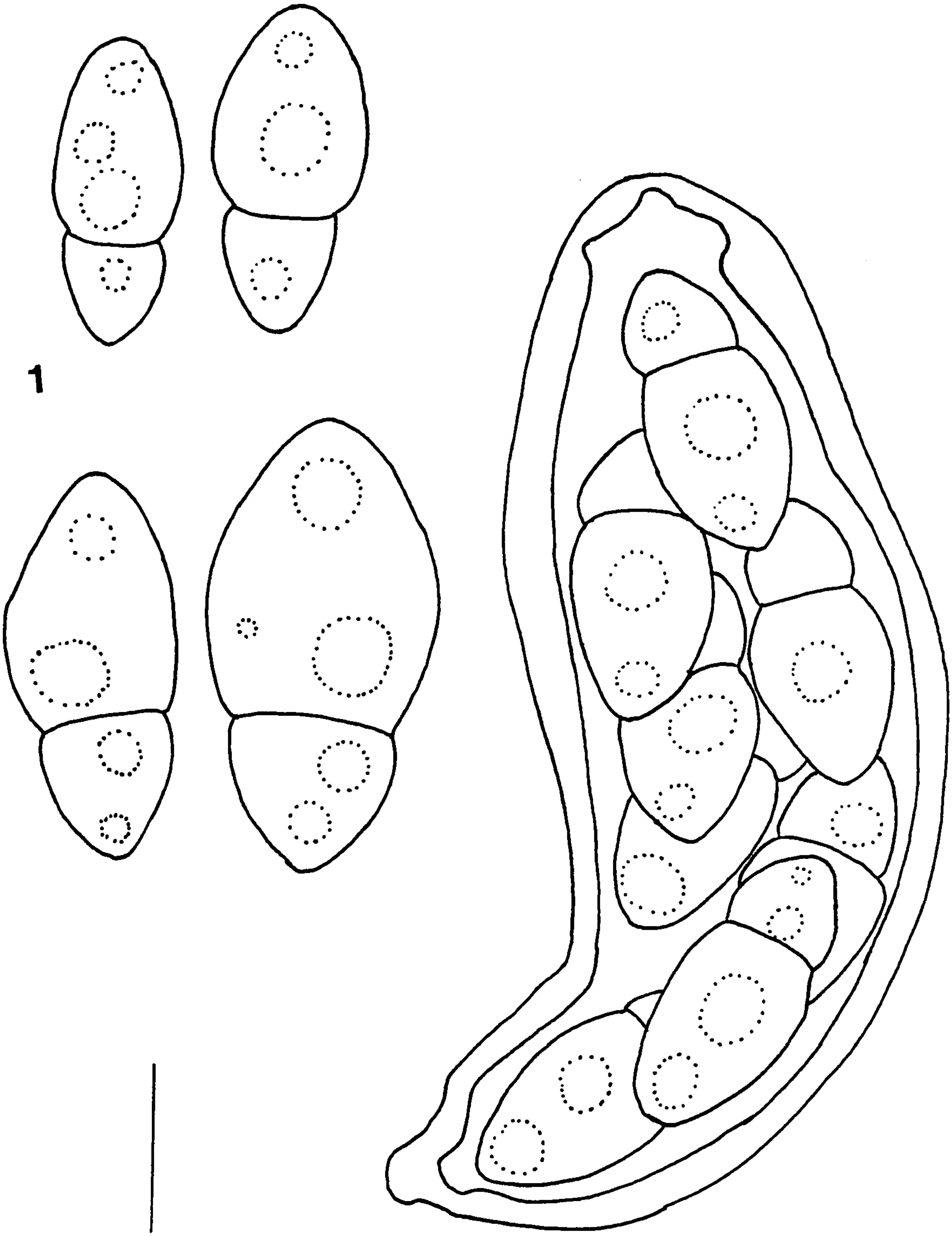
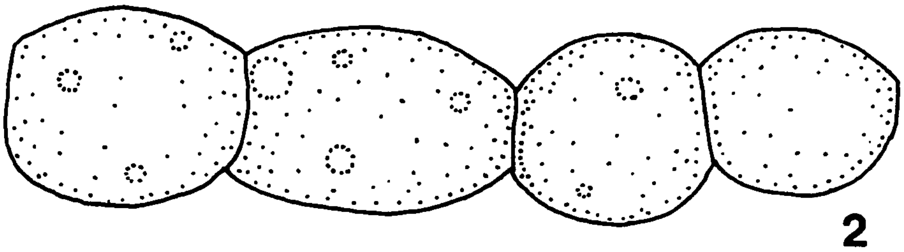

1
2

PLATE II

Figs. 1-3. *Calonectria* sp.

Fig. 1. Ascus.
Fig. 2. Chlamydospores.
Fig. 3. Ascospores.

Scale bar = 10 μm



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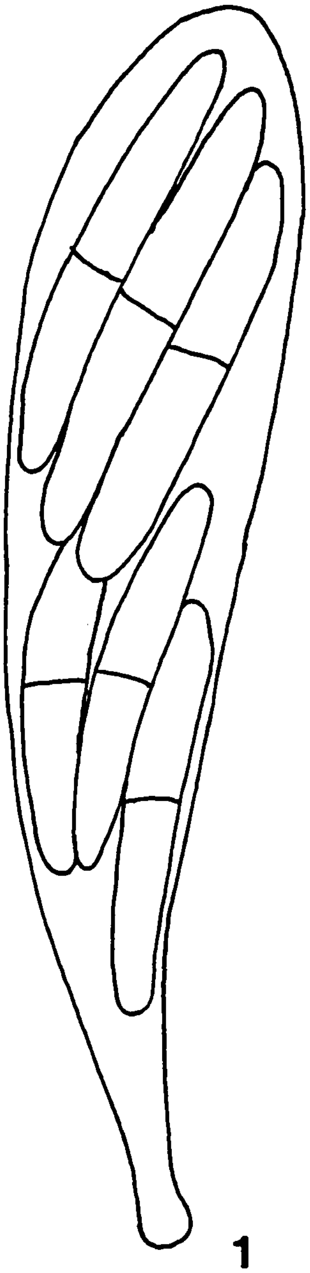

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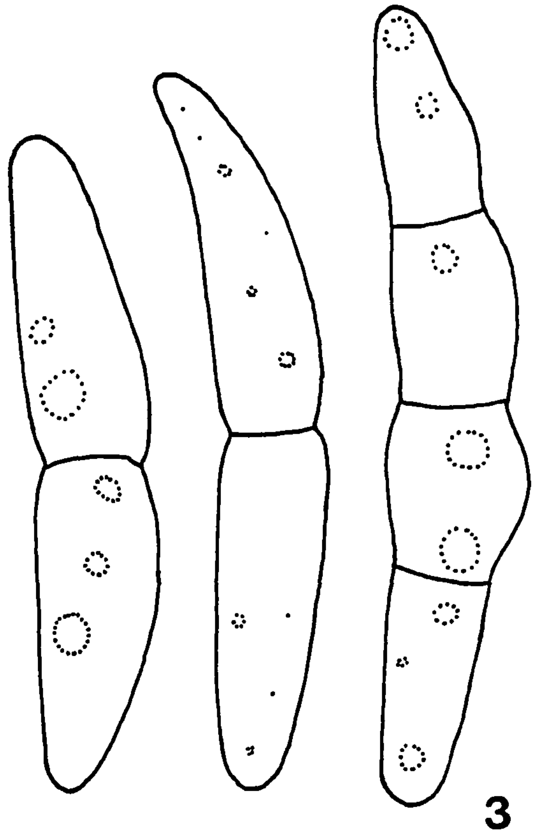


3

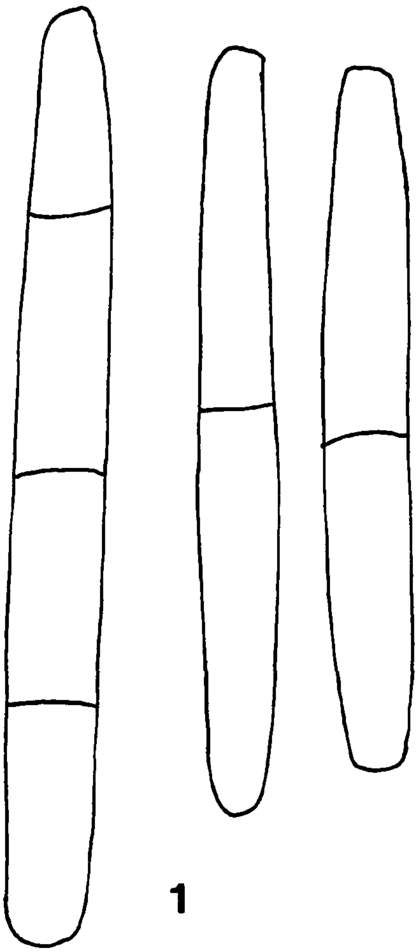
PLATE III

Figs. 1-2. *Cylindrocladium*

Fig. 1. Conidia.

Fig. 2. Conidiophore.

Scale bar = 10 µm



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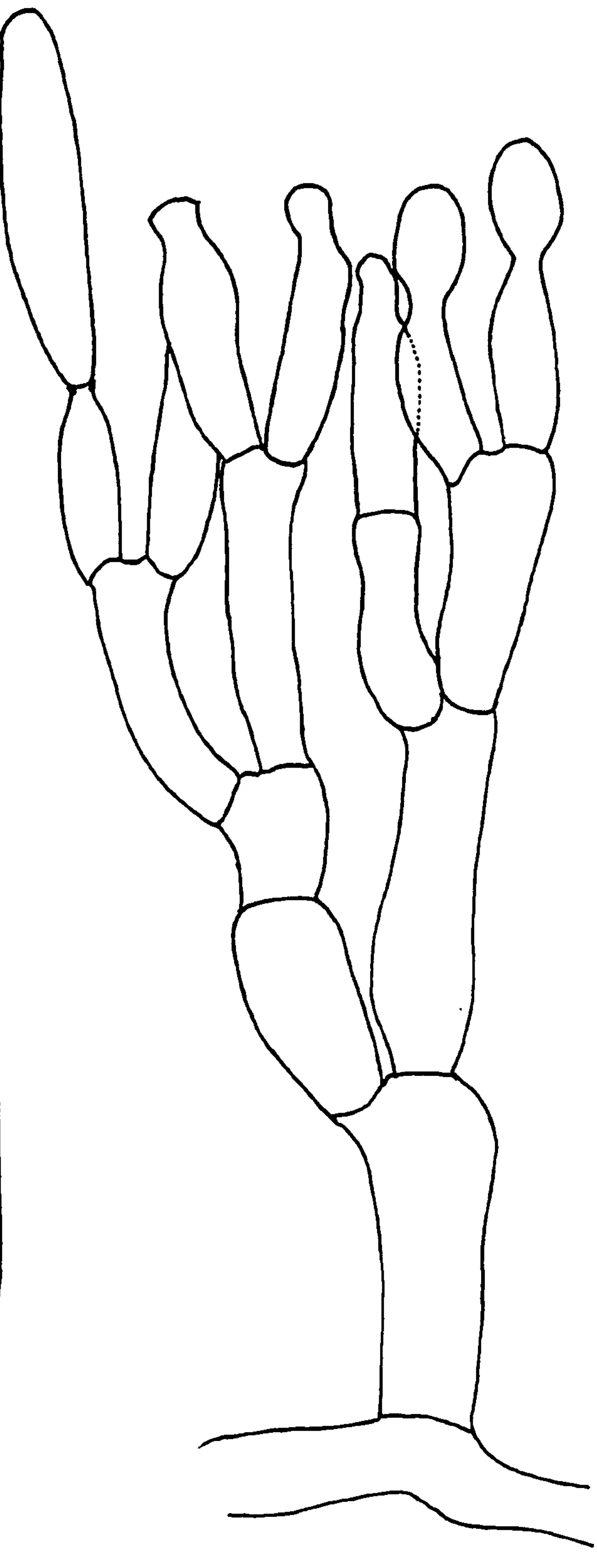


2

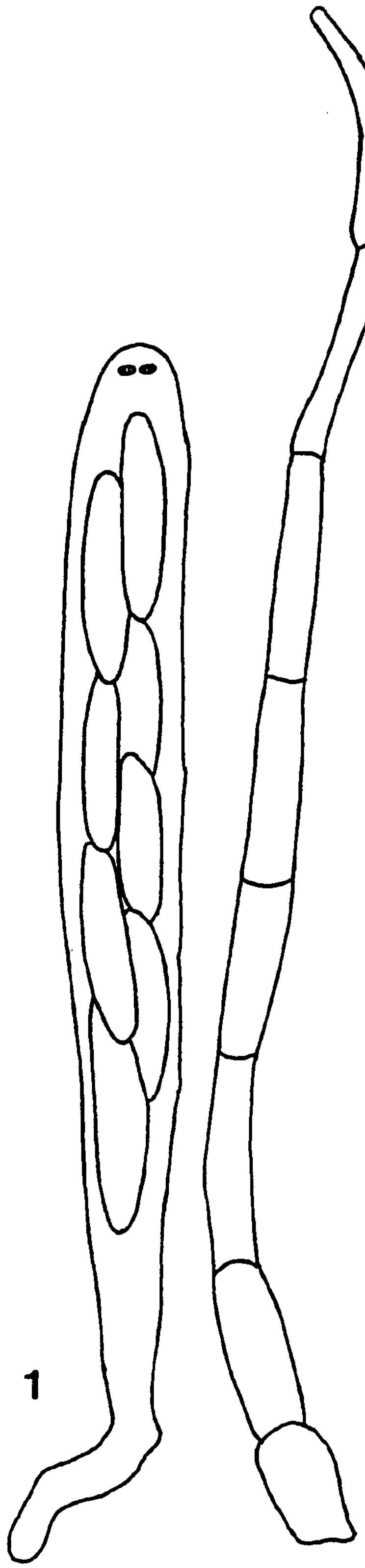
PLATE IV

Figs. 1-2. *Phomatospora* sp.

Fig. 1. Ascus and paraphyse, in Melzer's reagent.

Fig. 2. Ascospores.

Scale bar = 10 µm



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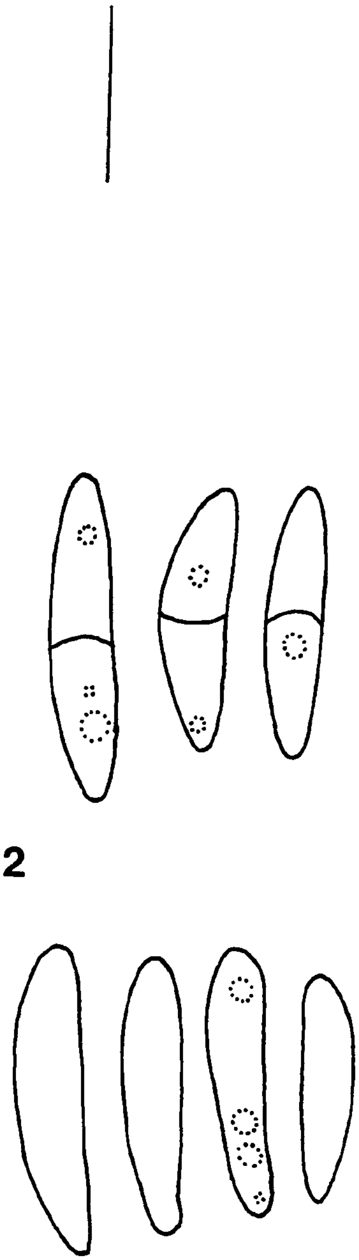


2

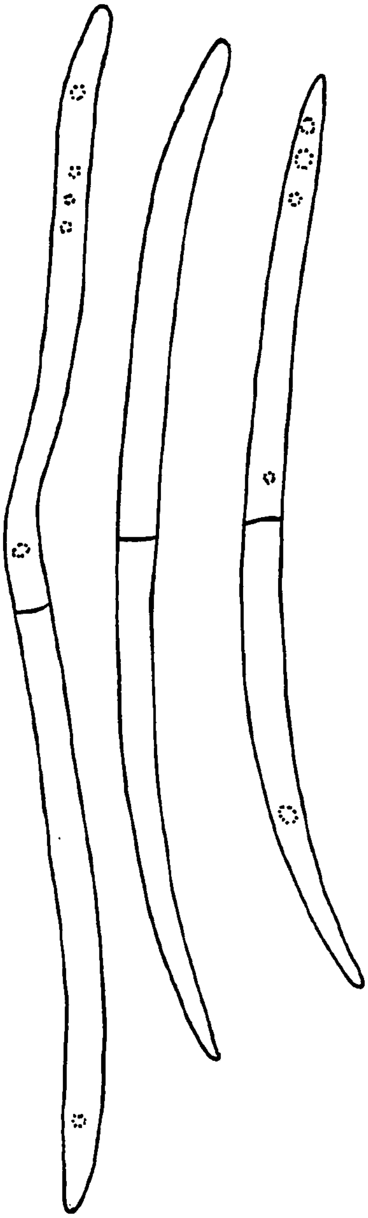
PLATE V

Figs. 1-2. *Leiosphaerella cocoes*

Fig. 1. Ascospores.

Fig. 2. Ascus tip, in Melzer's reagent.

Scale bar = 10 µm



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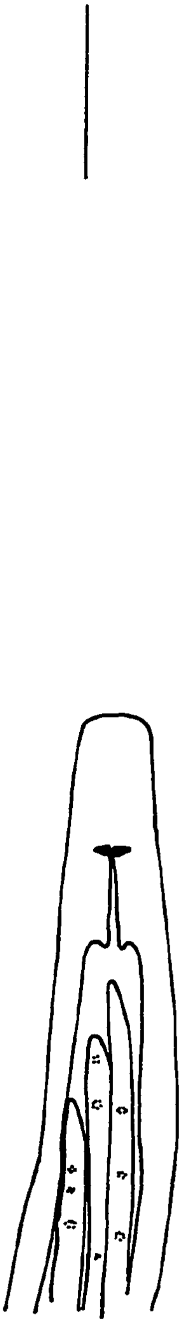


2

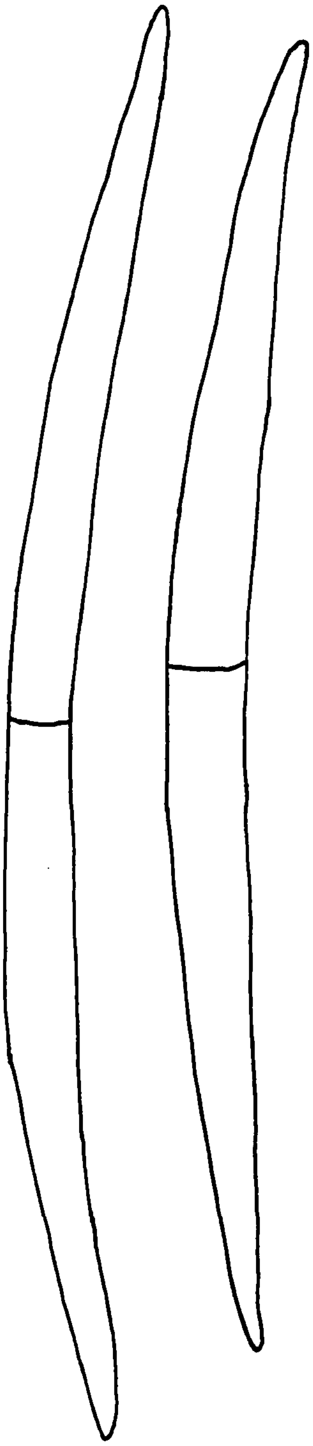
PLATE VI

Figs. 1-2. *Oxydothis poliothea*

Fig. 1. Ascospores.

Fig. 2. Ascus tip, in Melzer's reagent.

Scale bar = 10 µm



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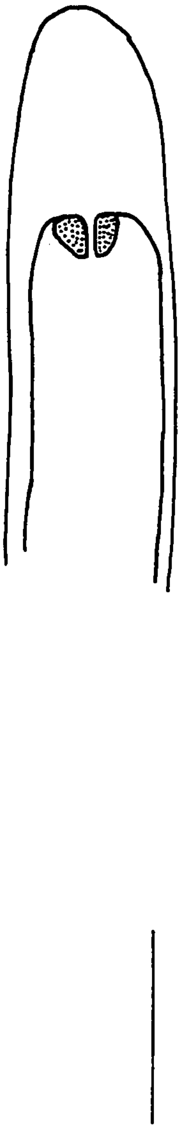


2

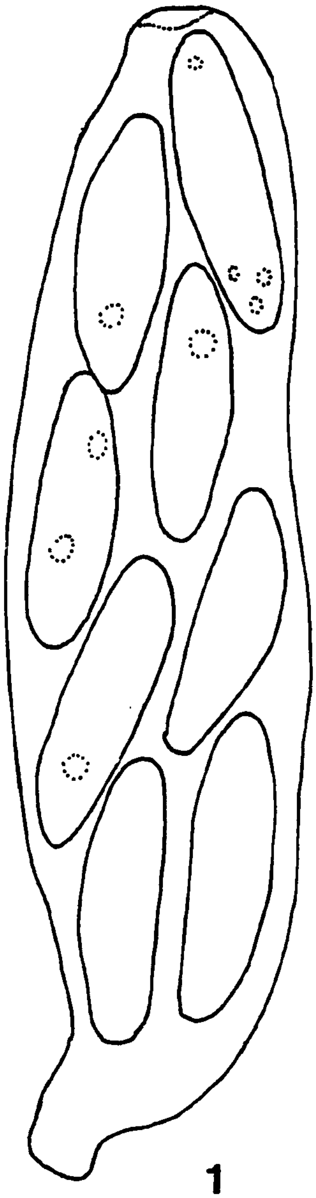
PLATE VII

Figs. 1-2. *Physalospora* sp.

Fig. 1. Ascus.

Fig. 2. Ascospores.

Scale bar = 10 μm



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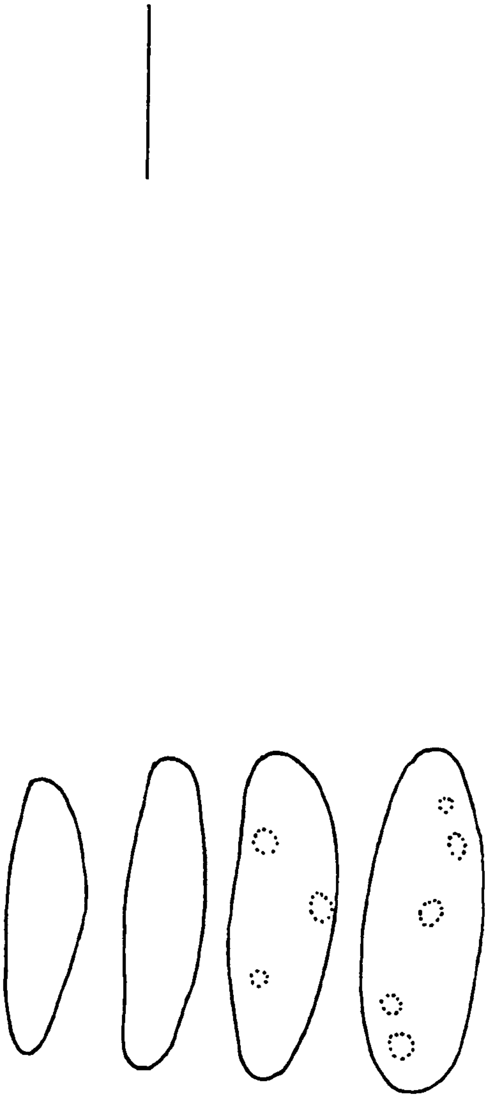

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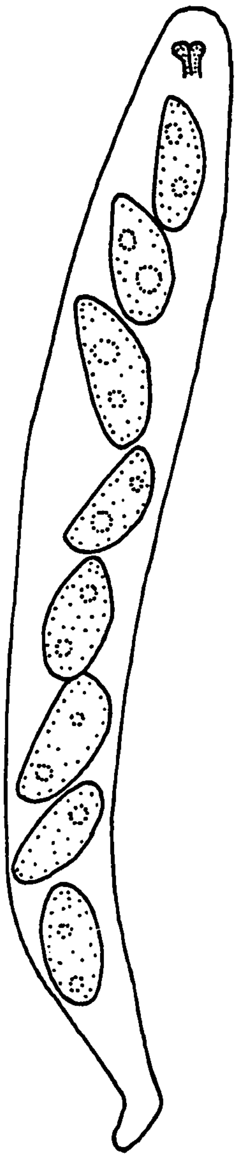
PLATE VIII

Figs. 1-2. *Anthostomella* sp.

Fig. 1. Ascus, and ascus tip, in Melzer's reagent.

Fig. 2. Ascospores.

Scale bar = 10 μm



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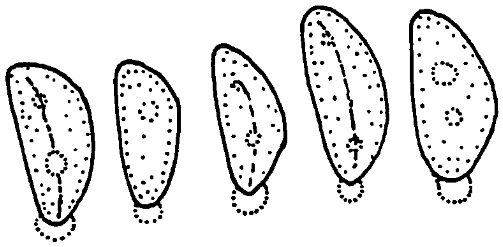

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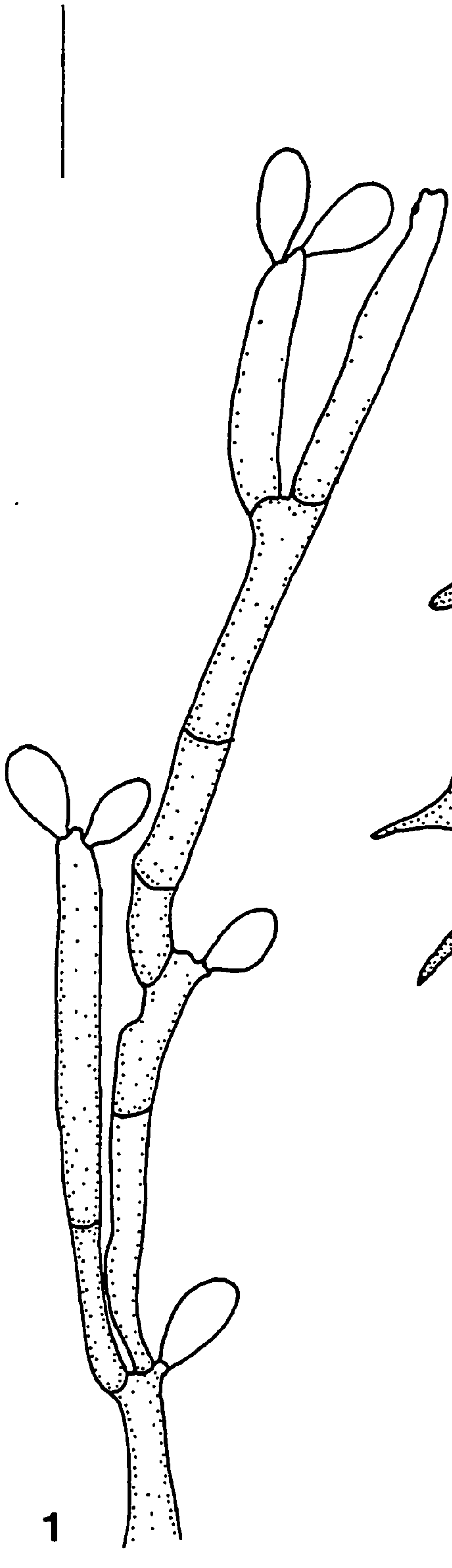
PLATE IX

Figs. 1-3. *Daldinia eschscholzii*

Fig. 1. Conidiophore.
Fig. 2. Conidia.
Fig. 3. Reticulated stromatic hyphae.

Scale bar = 10 µm



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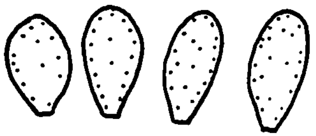

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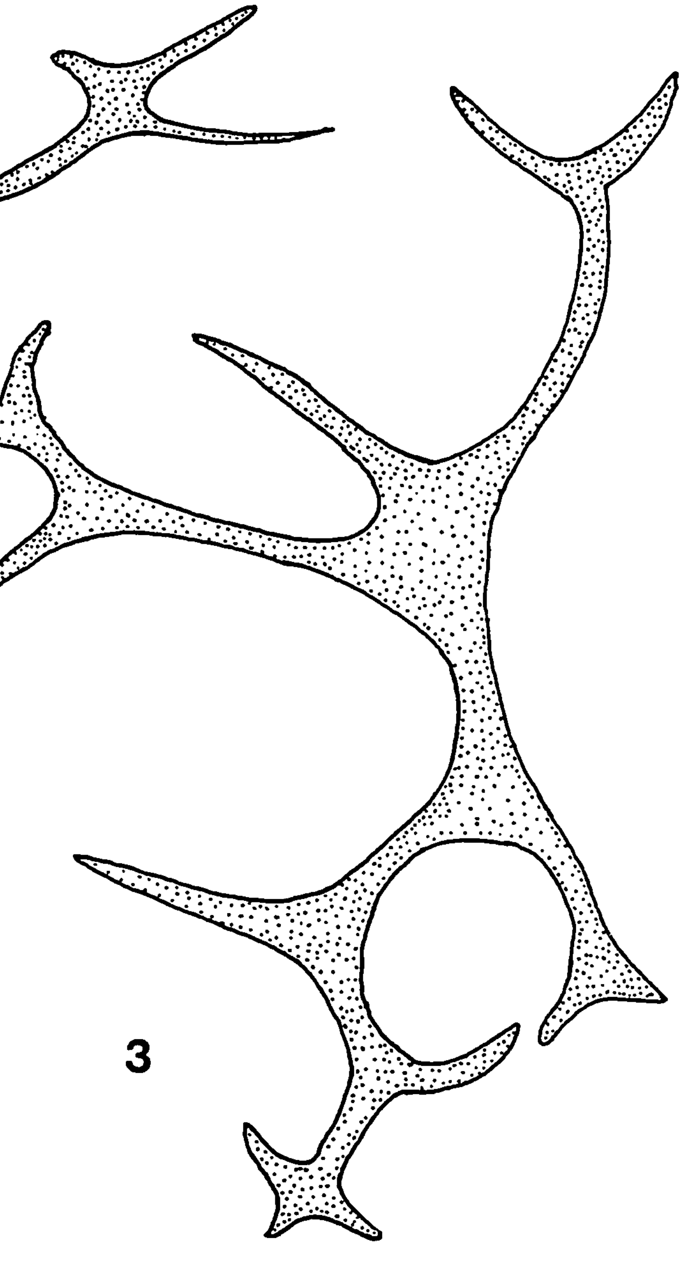

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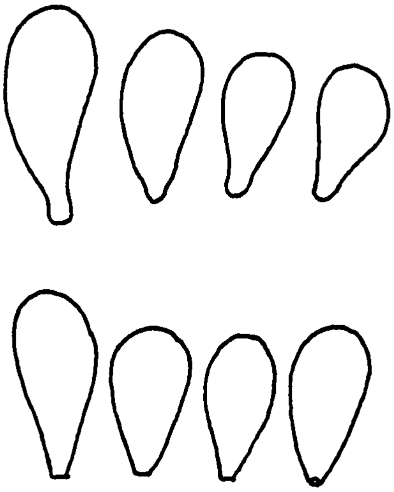
PLATE X

Figs. 1-3. *Hypoxylon quisquiliarum* (anamorph: *Nodulisporium*)

Fig. 1. Conidiophore.
Fig. 2. Conidia.
Fig. 3. Chlamydospores.

Scale bar = 10 μm



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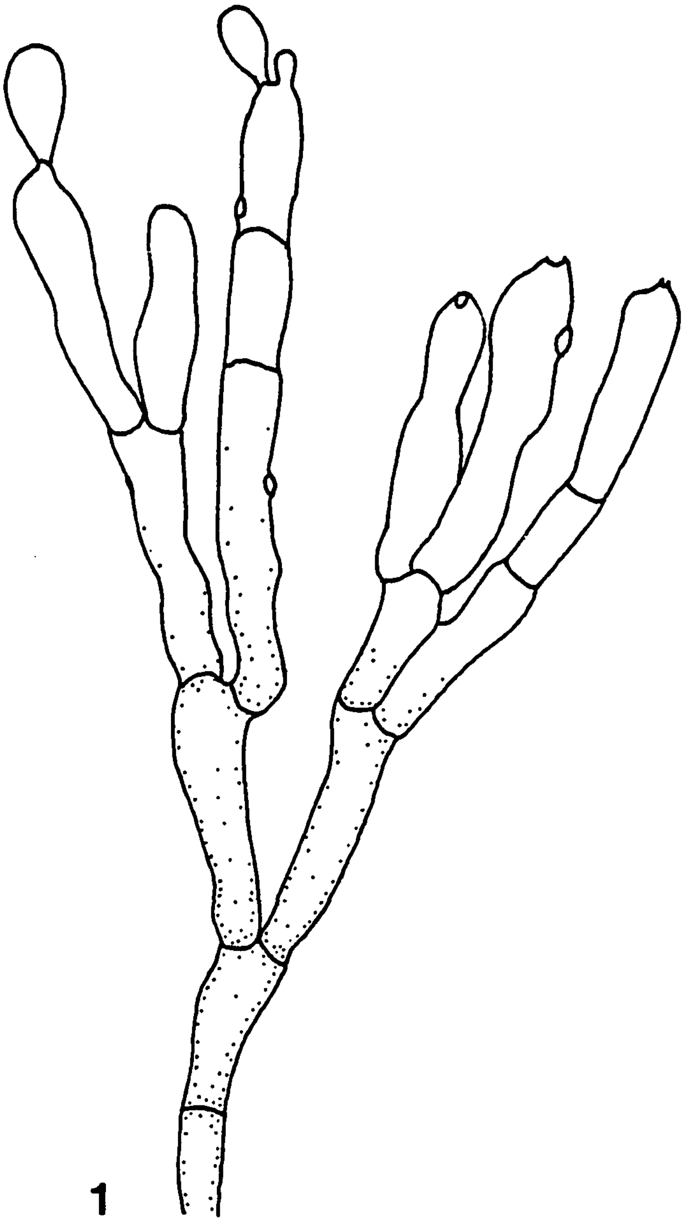

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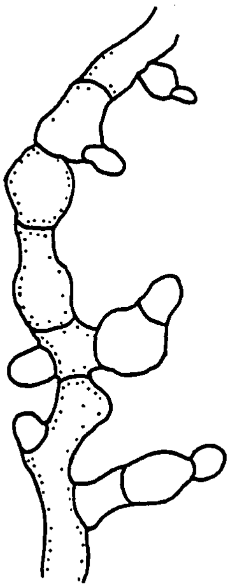

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PLATE XI

Figs. 1-3. *Hypoxylon serpens* (anamorph: *Geniculosporium*)

Fig. 1. Conidiophore.

Fig. 2. Conidia.

Fig. 3. Stromatic hyphae.

Scale bar = 10 µm

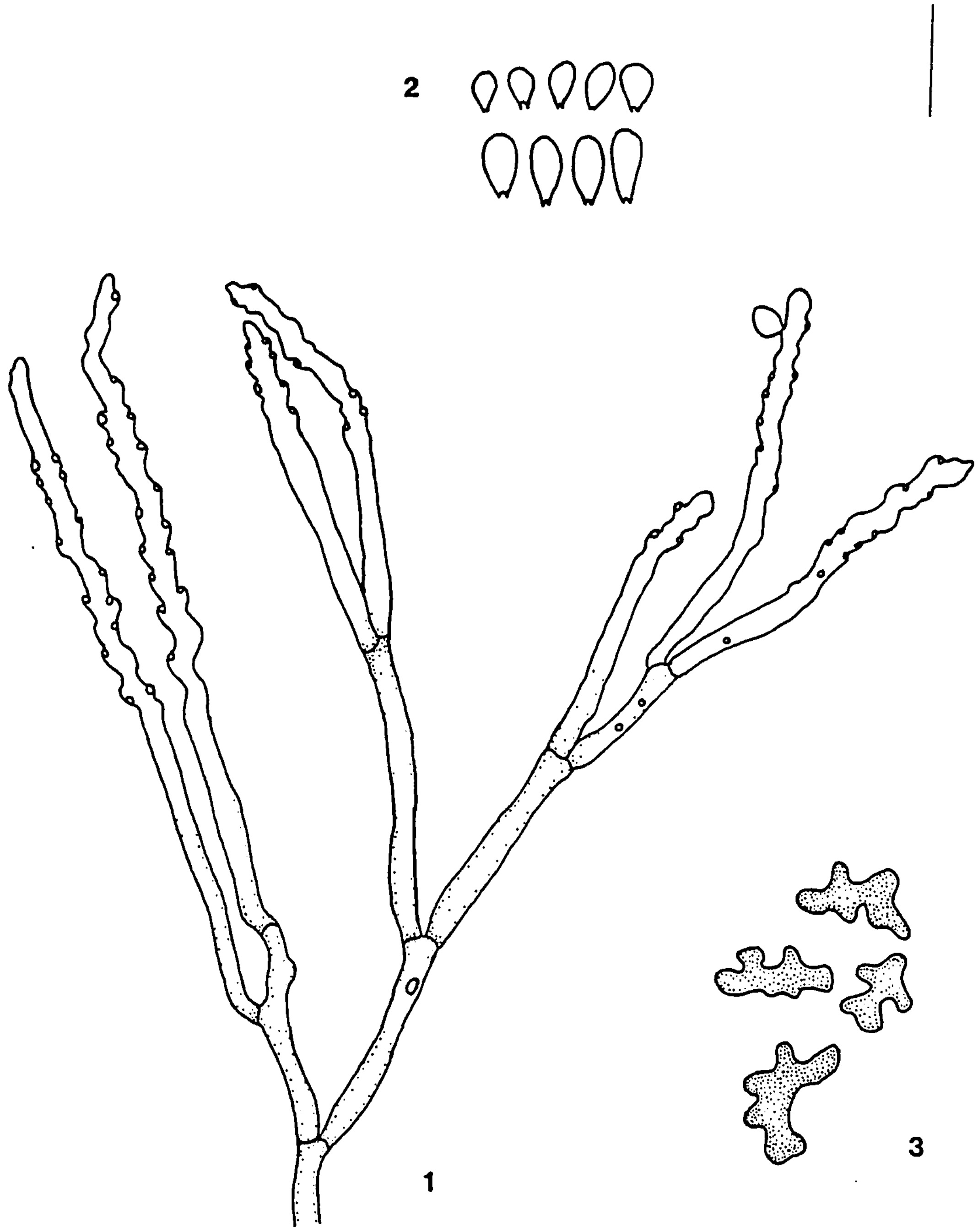
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PLATE XII

Figs. 1-2. *Hypoxylon stygium* (anamorph: *Nodulisporium*)

Fig. 1. Conidiophore.

Fig. 2. Conidia.

Scale bar = 10 µm

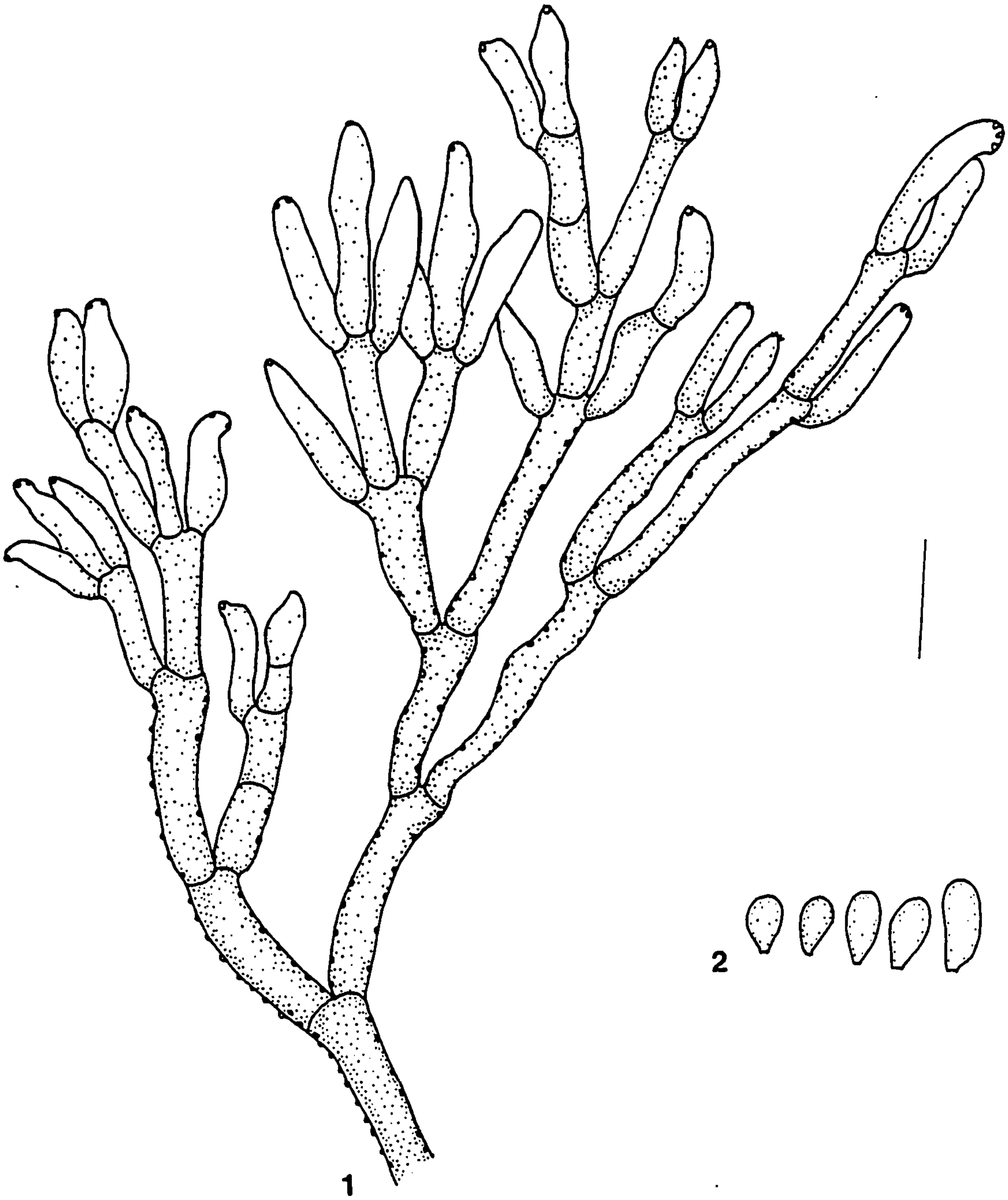
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PLATE XIII

Figs. 1-2. *Hypoxylon* sp. (anamorph: *Virgariella*)

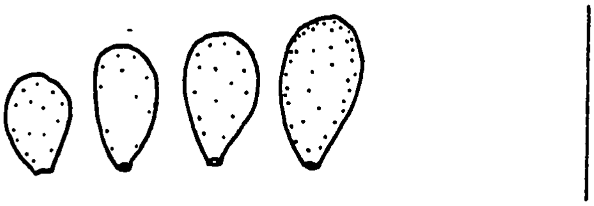
Fig. 1. Conidiophore.
Fig. 2. Conidia.

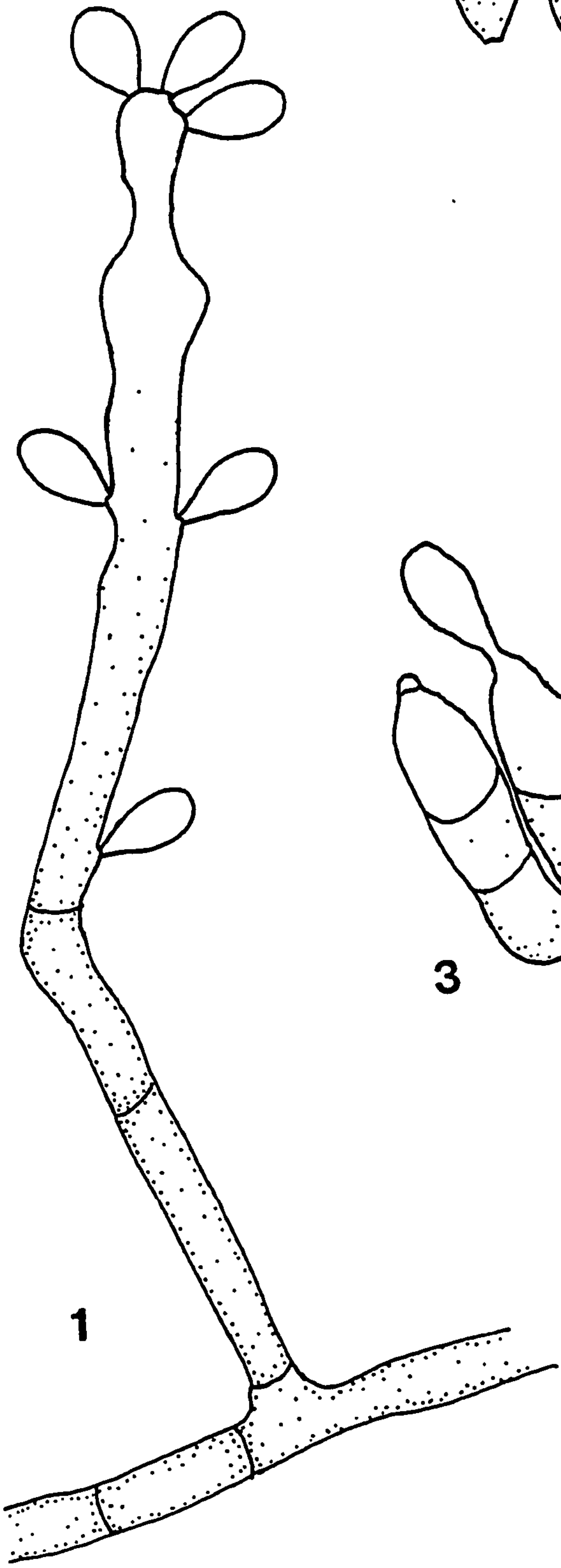
Figs. 3-4. *Ustulina ? deusta* (anamorph: *Nodulisporium*)

Fig. 3. Conidiophore.
Fig. 4. Conidia.

Scale bar = 10 µm

2





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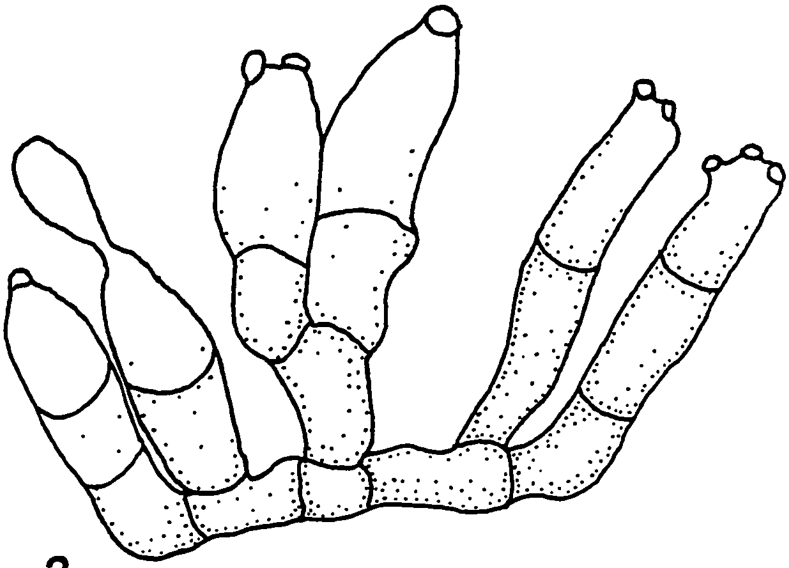

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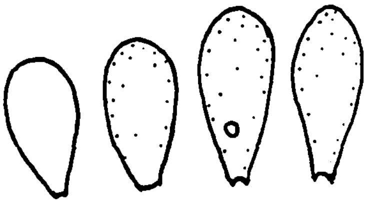

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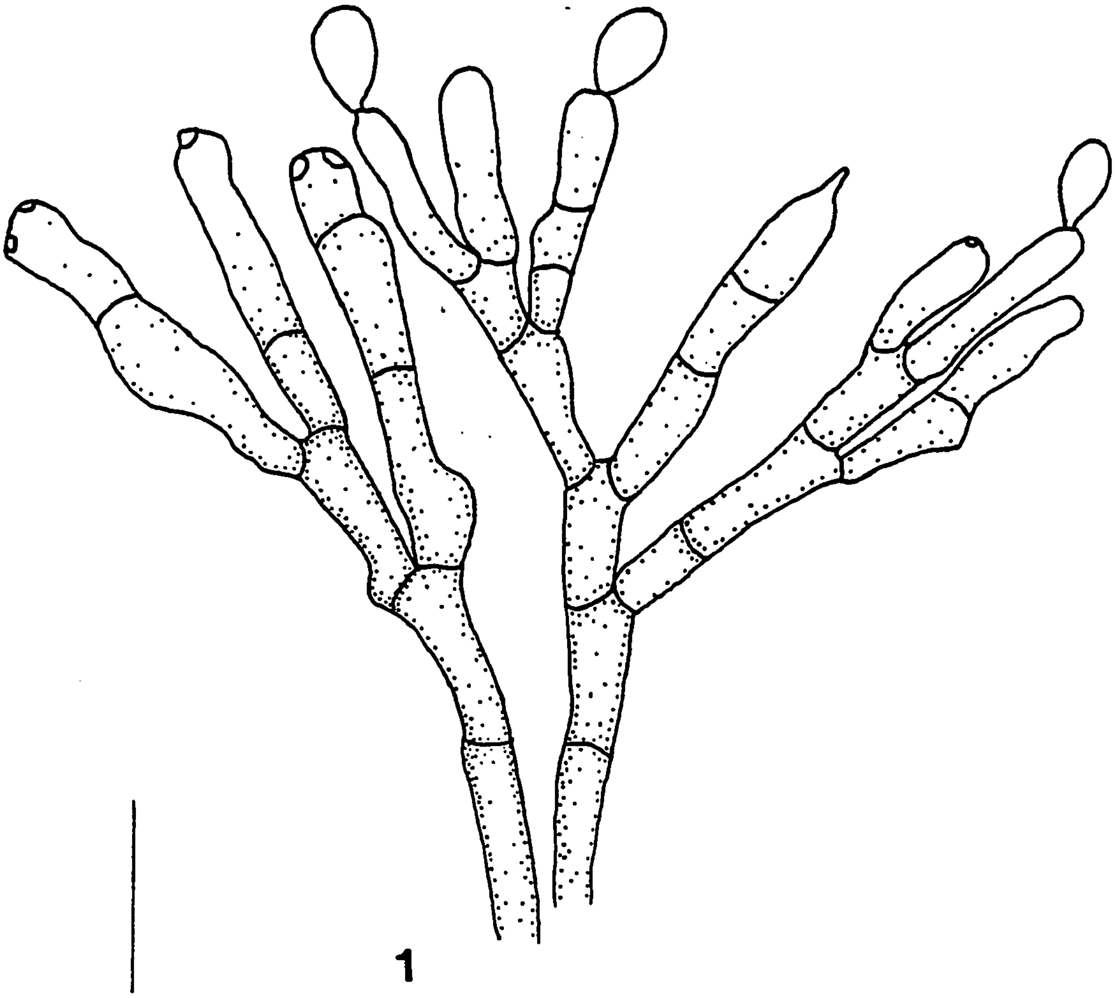
PLATE XIV

Figs. 1-3. *Nodulisporium* sp.

Fig. 1. Conidiophore.
Fig. 2. Conidia.
Fig. 3. Coiled stromatic hyphae.

Scale bar = 10 μm



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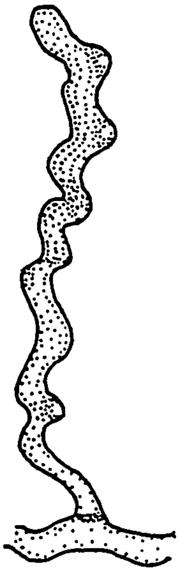

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PLATE XV

Figs. 1-2. *Penzigia* ? *indica* (anamorph: *Nodulisporium*)

Fig. 1. Conidiophores.

Fig. 2. Conidia.

Scale bar = 10 µm

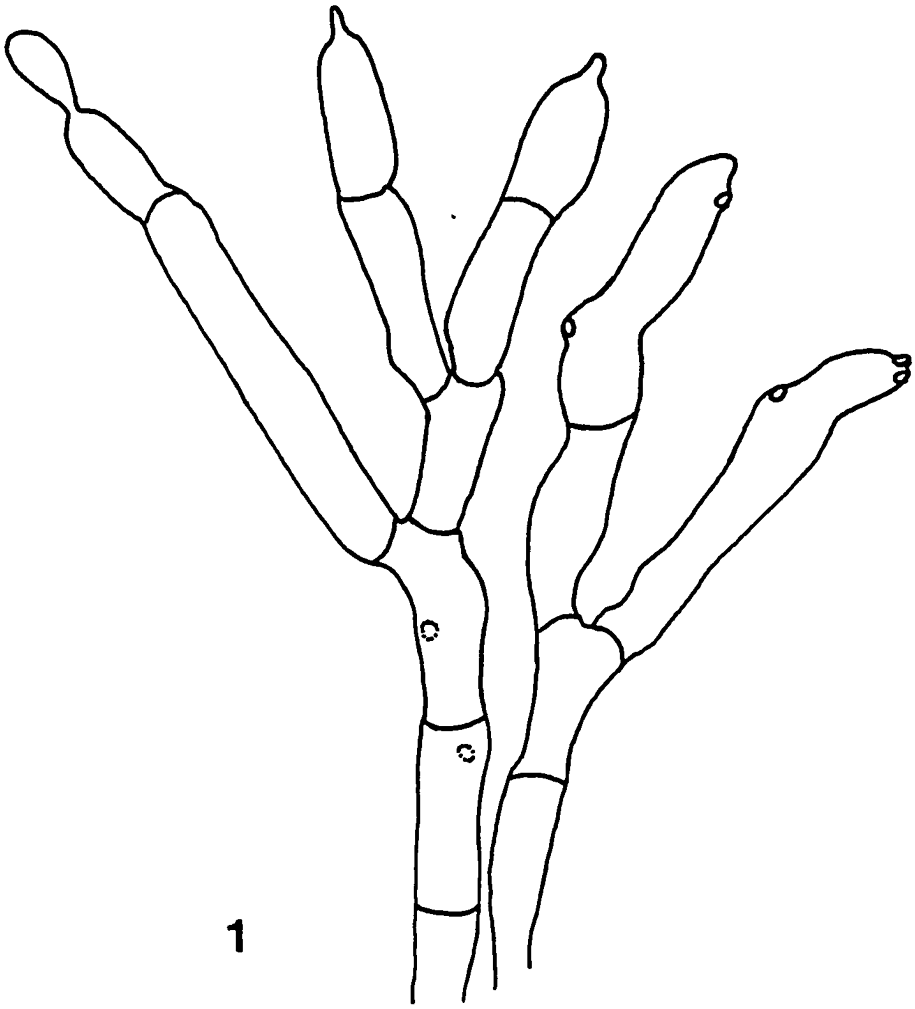

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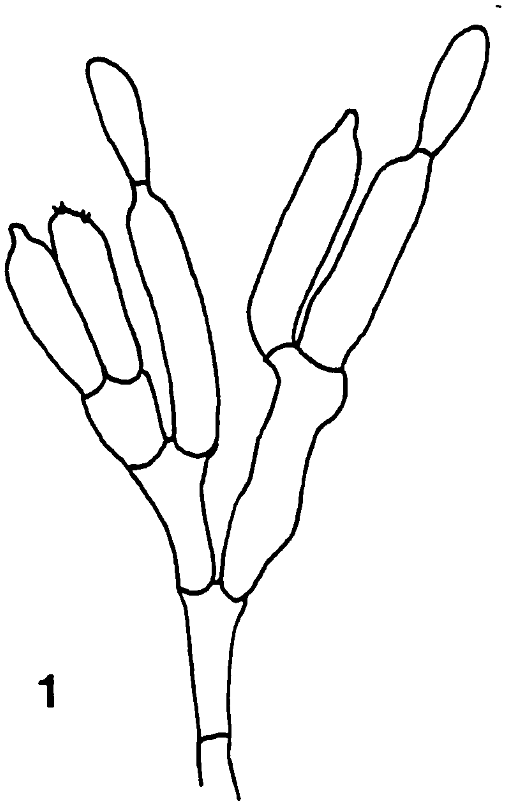
PLATE XVI

Figs. 1-4. *Xylaria adscendens* (anamorph: *Nodulisporium*)

Fig. 1. Conidiophore.
Fig. 2. Conidia.
Fig. 3. Segment of hyphae with warty excrescences.
Fig. 4. Segment of stromatic hyphae with short protuberances.

Scale bar = 10 μm



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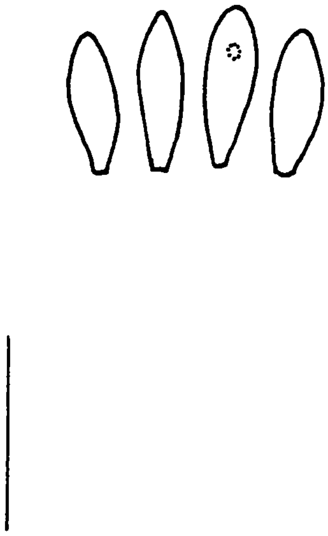

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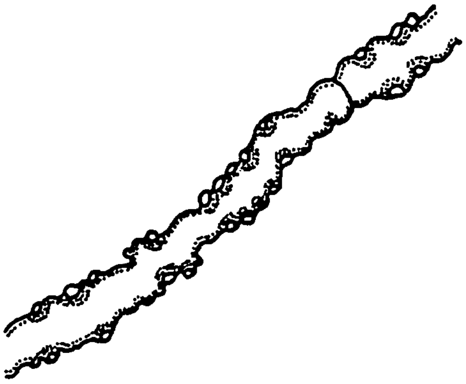

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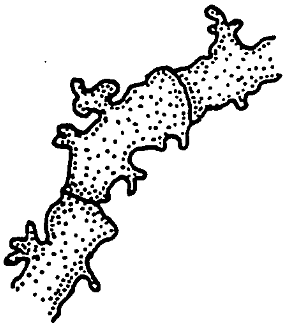


PLATE XVII

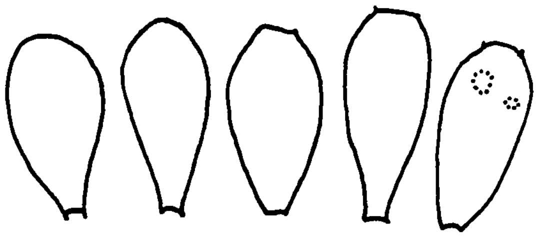
Figs. 1-2. *Xylaria allantoidea* (anamorph: *Nodulisporium*)

Fig. 1. Conidiophores.

Fig. 2. Conidia, with apical corona.

Scale bar = 10 µm

2



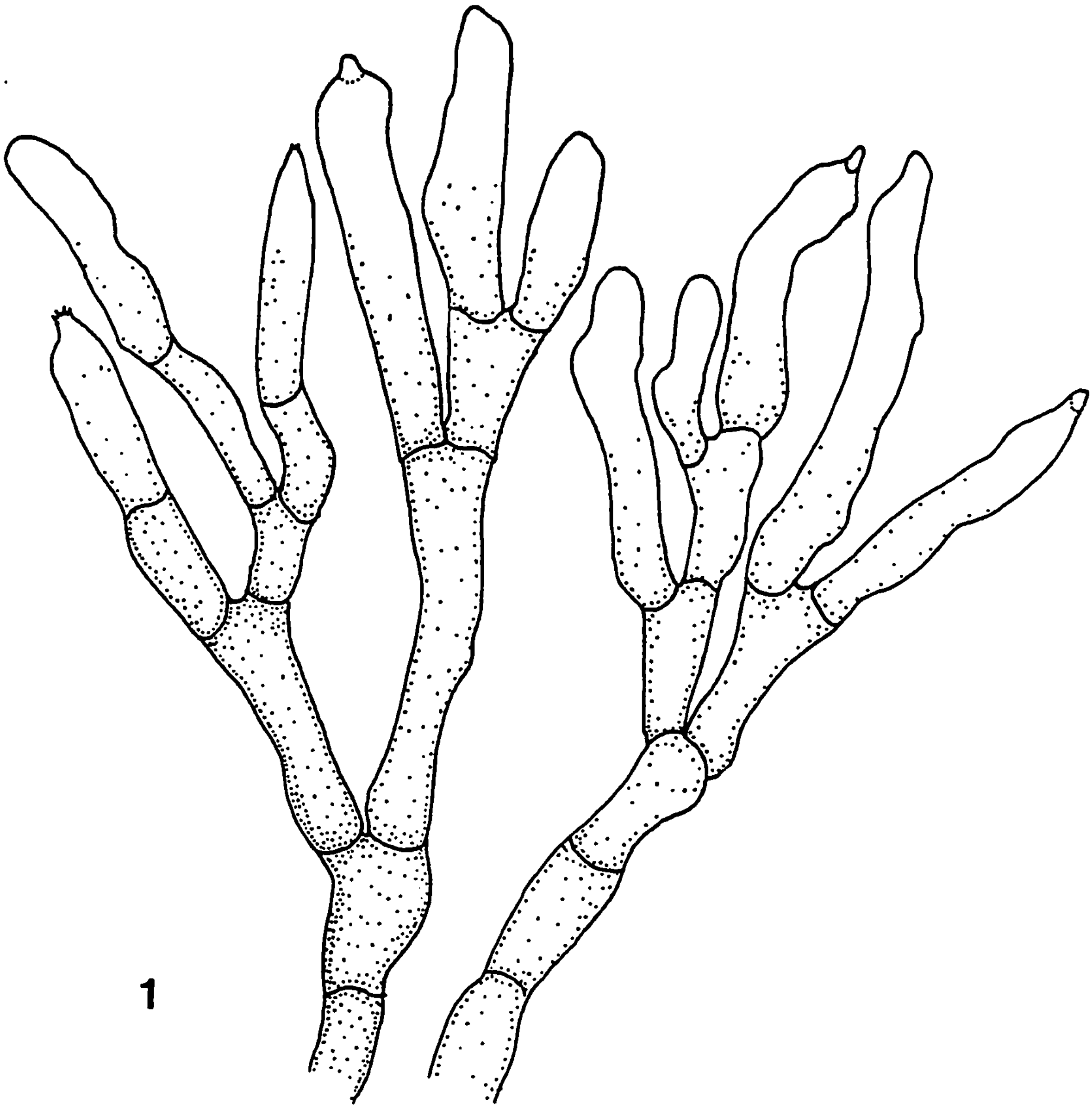


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PLATE XVIII

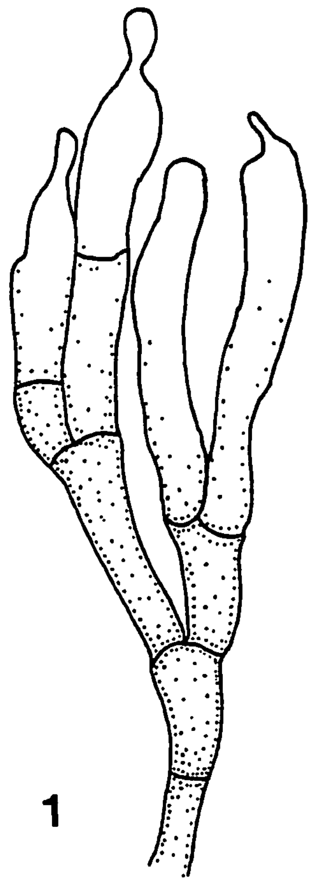
Figs. 1-3. *Xylaria anisopleura* (anamorph: *Nodulisporium*)

Fig. 1. Conidiophore.
Fig. 2. Conidia.
Fig. 3. Coiled stromatic hyphae.

Figs. 4-6. *Xylaria arbuscula* (anamorph: *Nodulisporium*)

Fig. 4. Conidiophore.
Fig. 5. Conidia.
Fig. 6. Long stromatic hyphae with protuberances.

Scale bar = 10 µm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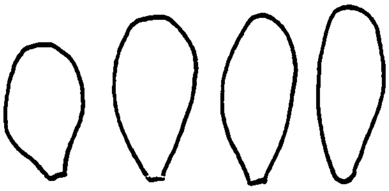
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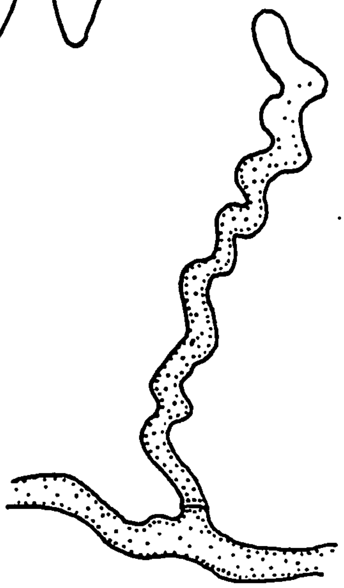
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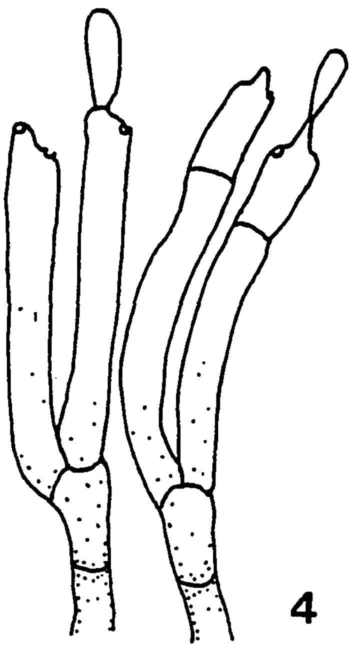
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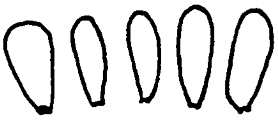
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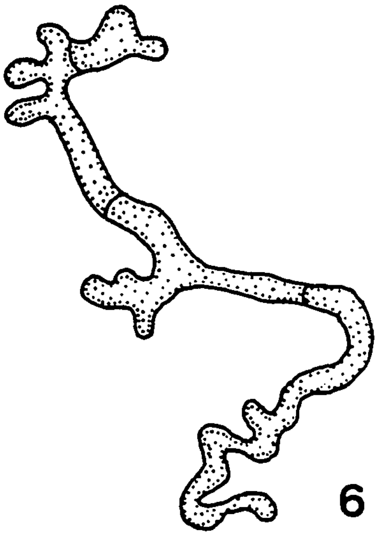
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PLATE XIX

Figs. 1-4. *Xylaria coccophora* (anamorph: *Nodulisporium*)

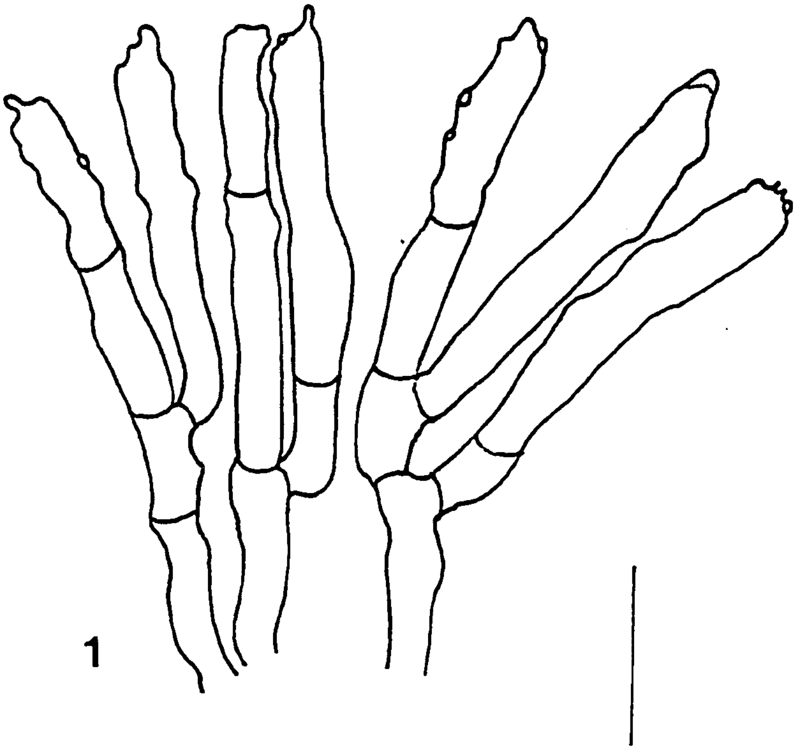
Fig. 1. Conidiophores.

Fig. 2. Conidia.

Fig. 3. Short stromatic hyphae.

Fig. 4. Segment of hyphae with warty excrescences.

Scale bar = 10 µm



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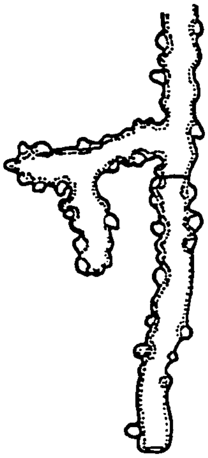

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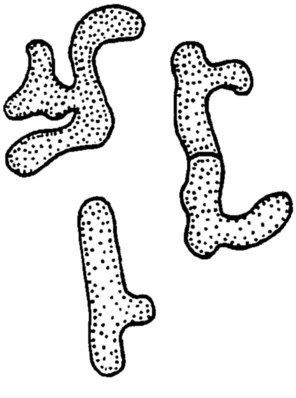

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PLATE XX

Figs. 1-2. *Xylaria cubensis* (anamorph: *Xylocoremium flabelliforme*)

Fig. 1. Conidiophores.
Fig. 2. Conidia.

Scale bar = 10 µm

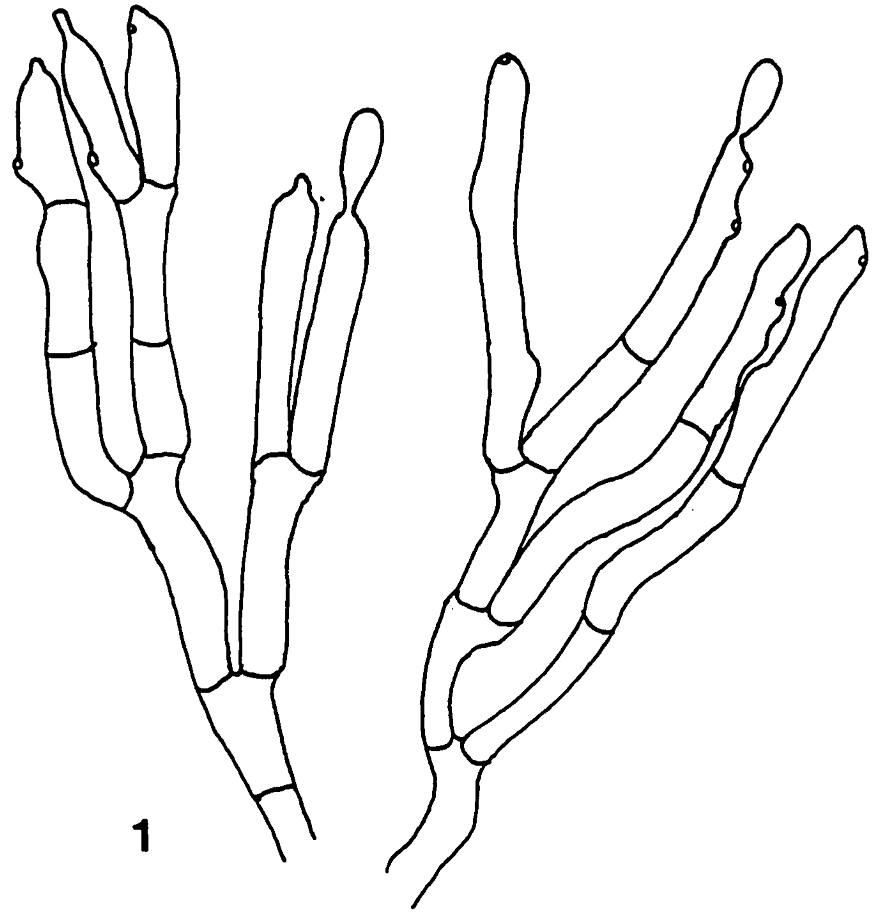

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PLATE XXI

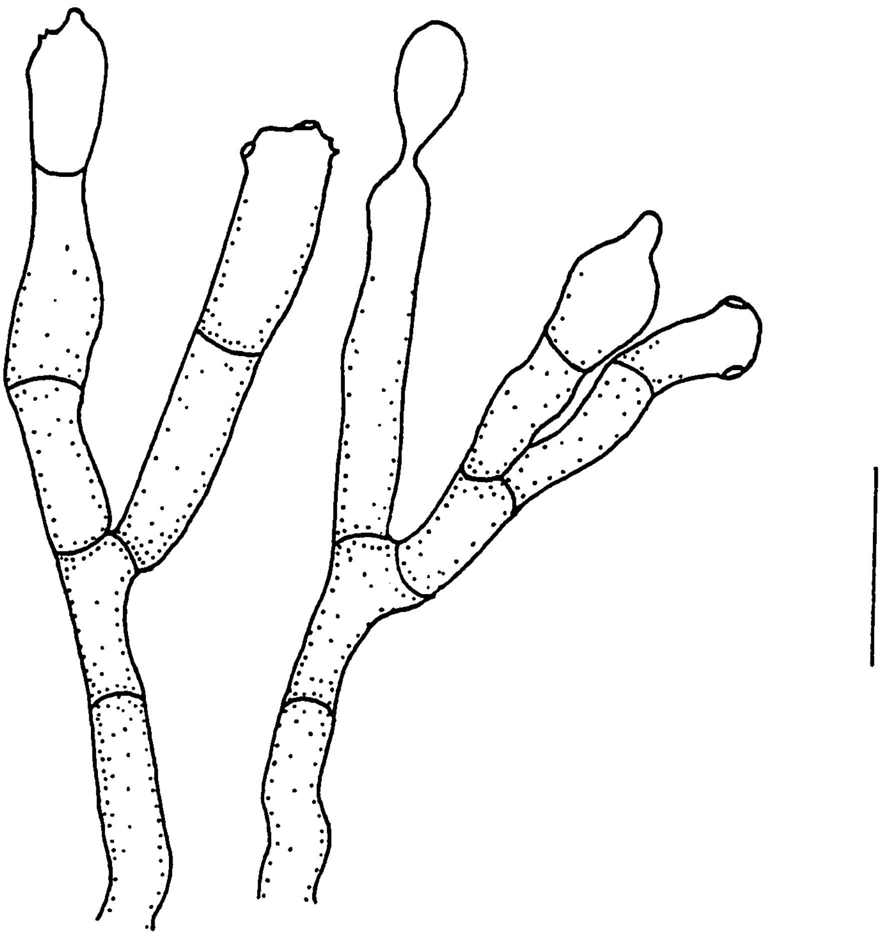
Figs. 1-3. *Xylaria curta* (anamorph: *Nodulisporium*)

Fig. 1. Conidiophores.

Fig. 2. Conidia.

Fig. 3. Stromatic hyphae with protuberances.

Scale bar = 10 µm



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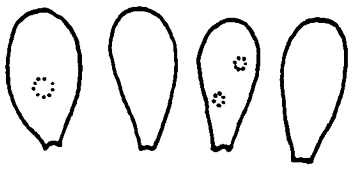

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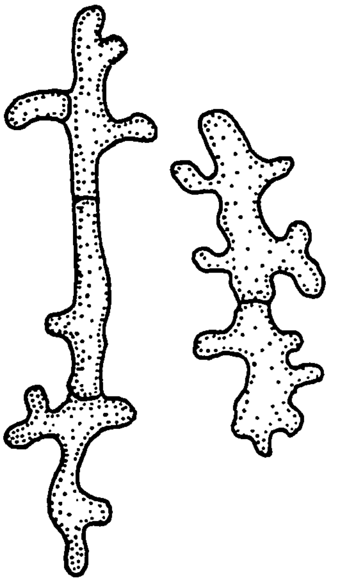

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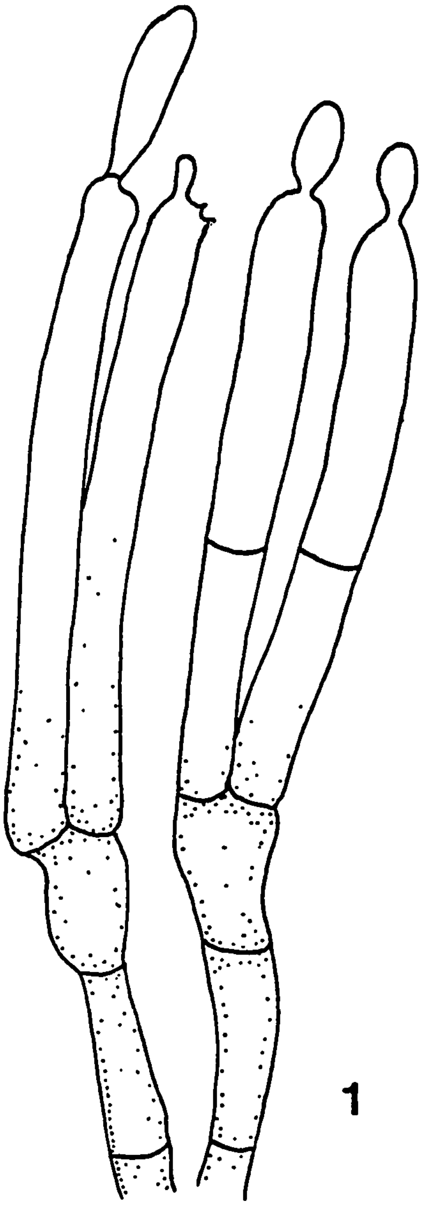
PLATE XXII

Figs. 1-3. *Xylaria obovata* (anamorph: *Nodulisporium*)

Fig. 1. Conidiophores.
Fig. 2. Stromatic hyphae with short protuberances.
Fig. 3. Conidia.

Scale bar = 10 μm



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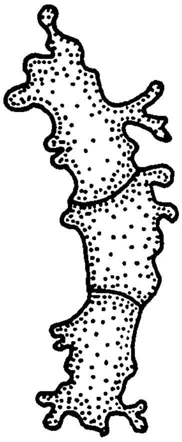

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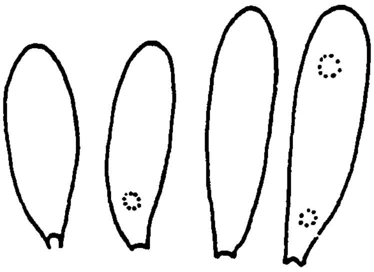

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PLATE XXIII

Figs. 1-2. *Acrodictys elaeidicola*

Fig. 1. Conidiophore.

Fig. 2. Conidia

Scale bar = 10 µm

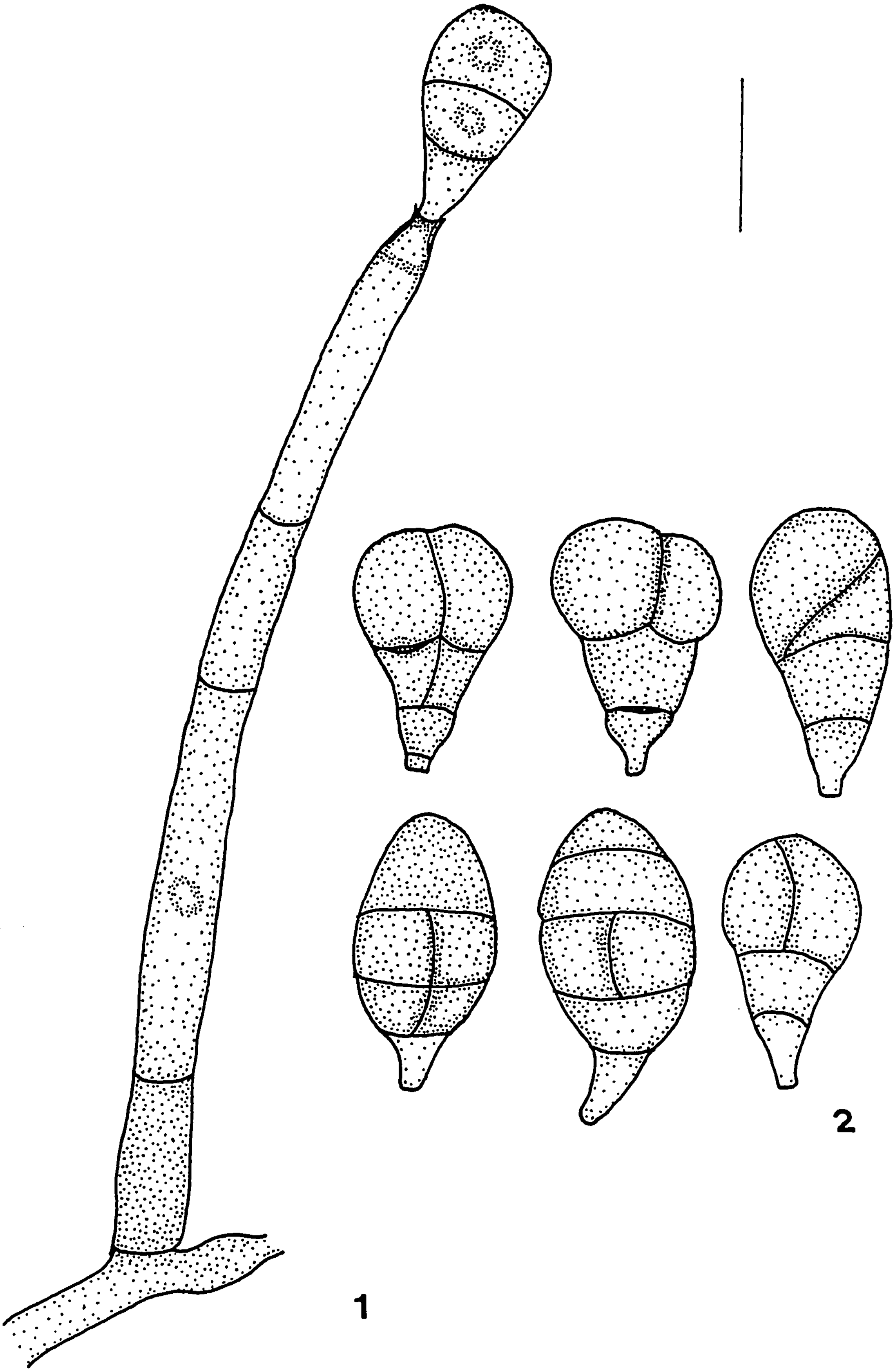

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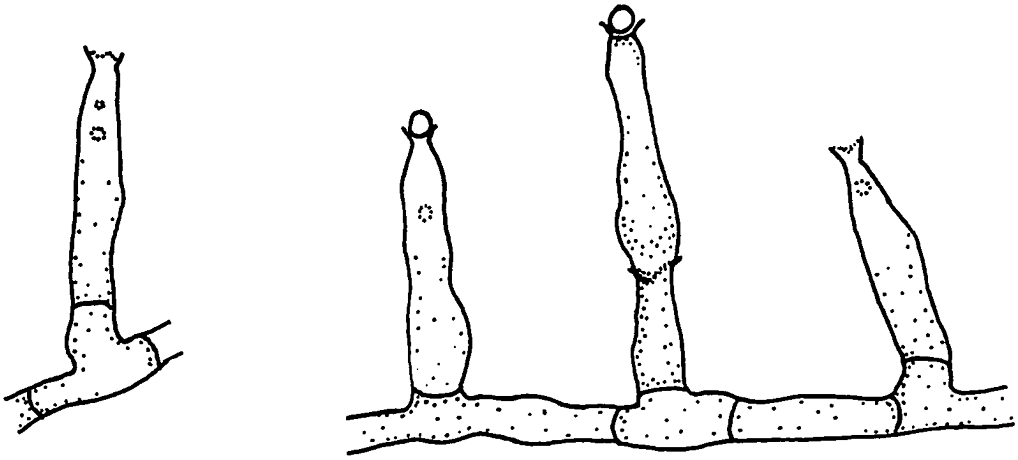
PLATE XXIV

Figs. 1-3. *Chloridium ? preussii*

Fig. 1. Conidiophores.
Fig. 2. Chlamydospores.
Fig. 3. Conidia.

Scale bar = 10 μm



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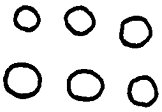

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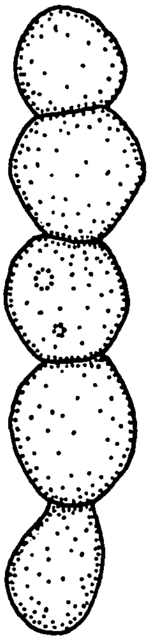

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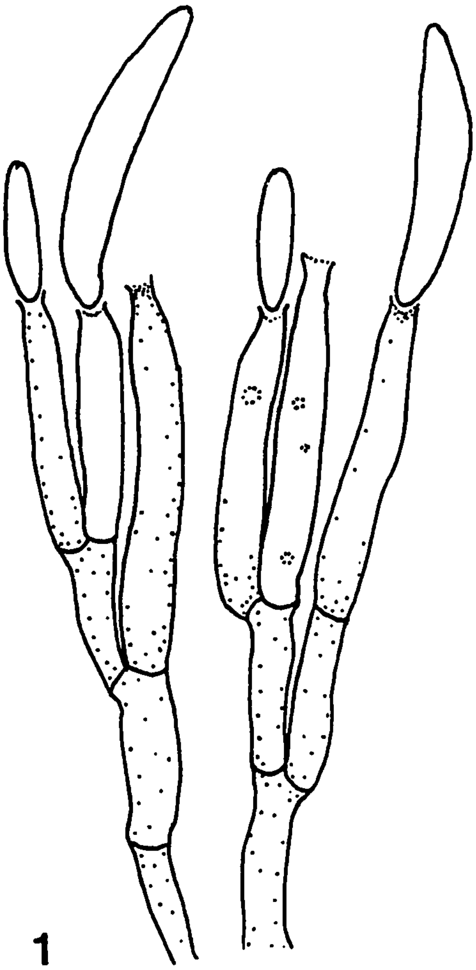
PLATE XXV

Figs. 1-3: *Thozetella* sp.

Fig. 1. Conidiophores.
Fig. 2. Conidia.
Fig. 3. Chlamydospores.

Scale bar = 10 µm



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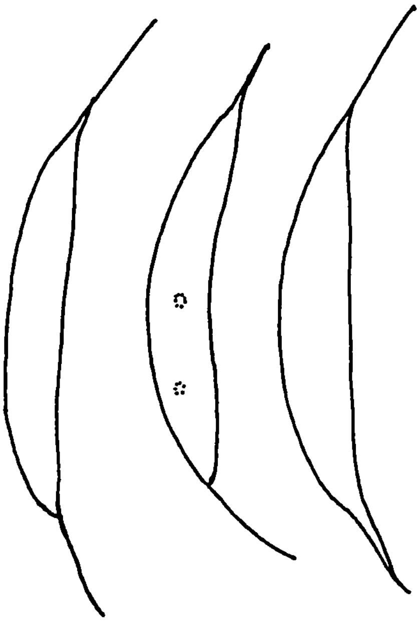

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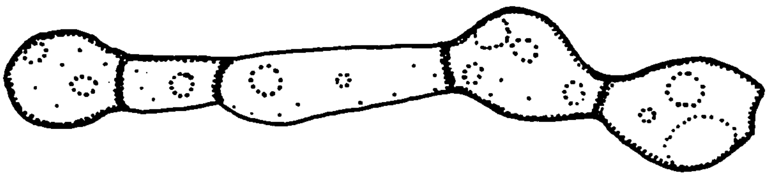

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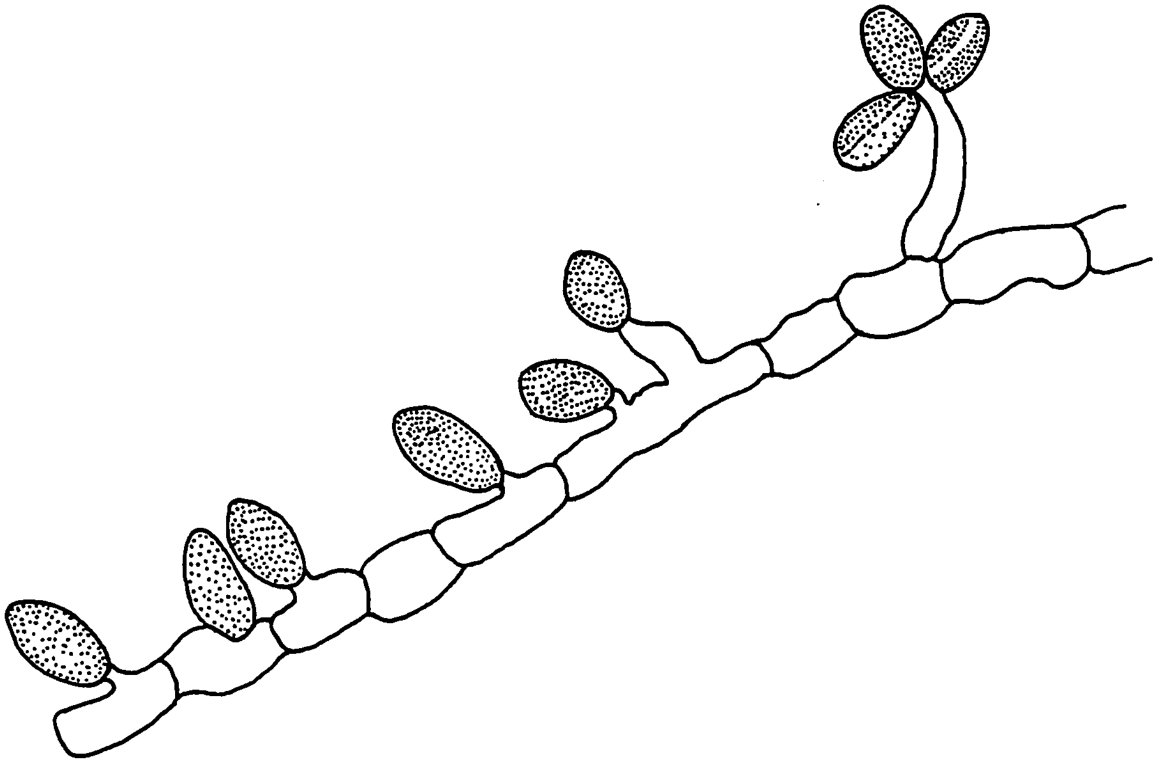
PLATE XXVI

Figs. 1-2: *Wardomyces* sp.

Fig. 1. Conidiophores.

Fig. 2. Conidia.

Scale bar = 10 µm



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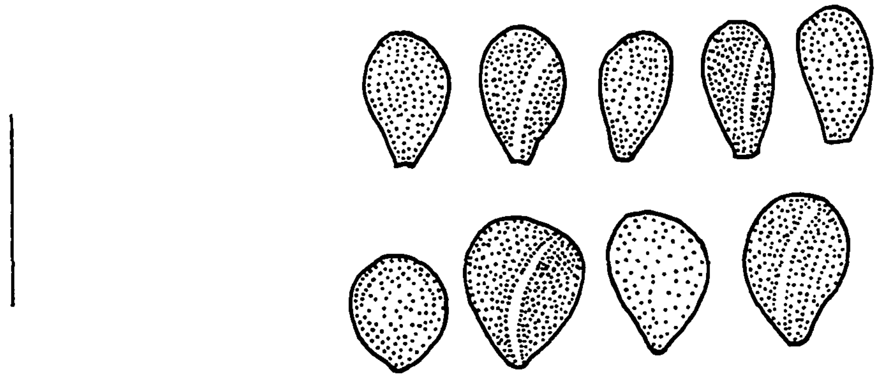

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APPENDIX 1

Culture media, stains and chemical substances

Culture media:

CYM liquid medium

KH_2PO_4	0.69 g
K_2HPO_4	1.50 g
$MgSO_4'7H_2O$	0.75 g
Dextrose	30 g
Yeast extract	3 g
Peptone	3 g
Distilled water	1.5 l

Oatmeal agar (OA)

Oatmeal	30 g
Bacto agar	12.5 g
Distilled water	500 ml

Obs: blended for 2 minutes, then autoclaved.

Phytone - yeast extract agar (PYE)

Phytone	10 g
Dextrose	40 g
Yeast extract	5 g
Agar	20 g
Distilled water	1 l

SNA

KH_2PO_4	1 g
KNO_3	1 g
$MgSO_4.7H_2O$	0.5 g
KCl	0.5 g
Glucose	0.2 g
Saccharose	0.2 g
Distilled water	1 l
Agar-Agar	17-20 g

APPENDIX 1 (cont.)

V 8 liquid medium

centrifuged V8 juice	100 ml
KH_2PO_4	0.5 g
Na_2HPO_4	0.5 g
$MgSO_4.7H_2O$	0.5 g
KCl	0.25 g
L-asparagin monohydrate	2 g
D-glucose	10 g
$FeCl_3$	0.01 g

Filled up to 1 l with distilled water.
pH 7.0

Stains and chemical substances:

Amann's Lactophenol

Phenol	20 g
Lactic acid	20 g
Glycerine	40 g
Distilled water	20 ml

Cotton Blue in Lactic Acid

Lactic acid (concentrate)	100 ml
Cotton blue	1.17 g

Obs: Gently heat after application of the cover glass.

Melzer's Reagent

Iodine	0.5 g
Potassium iodine	1.5 g
Chloral hydrate	20.0 g
Distilled water	20.0 ml

Cyclosporine A (CsA) stock solution: 20 ml

EtOH 95%	0.5 ml
Tween 80	0.2 ml
Sterile water	19.3 ml

Obs: store at room temperature

Streptomycin Sulfate: stock solution: 50 ml

Streptomycin	0.2 g
Sterile water	50 ml

Obs: store in the refrigerator

APPENDIX 2.

Buffers and enzyme recipes

Buffers:

Extraction buffer (Soltis et al. 1983):

Stock buffer	25 ml
PVP-40 (Polyvinyl-pyrrolidone)	2.5 g
2-mercaptoethanol	25 µl

Stock:

0.1 M Tris-HCl, pH 7.5	200 ml
$EDTA.Na_4$ dihydrate	76 mg
KCl	148 mg
$MgCl_2$, hexahydrate	400 mg

Buffer system I:

<u>Electrode buffer</u>:

citric acid trisodium dihydrate	235.28 g
H_2O distilled	2 l
pH 7.0	

<u>Gel buffer</u>:

L-Histidine.HCl monohydrate	8.38g
H_2O distilled	2 l
pH 7.0	

Buffer system II:

<u>Electrode buffer</u>

Tris	32.7 g
Citric acid	12.2 g
H_2O distilled	2 l
pH 8.0	

<u>Gel buffer</u>

dilute 30.2 ml electrode buffer to	450 ml
pH 8.0	

Buffer system III:

<u>Electrode buffer</u>:

Tris	54.0 g
Citric acid (anhydridous)	26.66 g
H_2O distilled	2 l
pH 7.2	

<u>Gel buffer</u>:

dilute	15.75 ml
electrode buffer	
to	450 ml
pH 7.2	

APPENDIX 2 (cont.)

Enzymes:

Aconitase (ACO)

0.1 M Tris, pH unadjusted	50 ml
cis-Aconitic acid	50 mg
Adjust pH to 8.0	
1.0 M $MgCl_2$	0.5 ml
NADP	5 mg
MTT	5 mg
PMS	1 mg
IDH	40 units

obs: stain in the dark at 37°C until bands appear. Rinse in water.

Aldolase (ALD)

0.1 M Tris-HCl, pH 8.5	50 ml
Fructose-1,6-diphosphate.Na_2	125 mg
Arsenic acid.Na_2	150 mg
Glyceraldehyd-3-phosphate dehydrogenase	100 units
NAD	20 mg
MTT	5 mg
PMS	1 mg

obs:stain in the dark at room temperature until bands appear. Rinse in water.

Malate dehydrogenase (MDH)

0.1 M Tris-HCl, pH 8.0	45 ml
2.0 M DL-malic acid, pH 8.0	5 ml
NAD	10 mg
MTT	5 mg
PMS	1 mg

obs: stain in the dark at room temperature until bands appear. Rinse in water.

Phosphoglucoisomerase (PGI)

0.1 M Tris-HCl, pH 8.0	50 ml
1.0 M $MgCl_2$	0.5 ml
Fructose-6-phosphate, disodium salt	15 mg
Glucose-6-phosphate dehydrogenase	80 units
NAD	5 mg
MTT	5 mg
PMS	1 mg

obs: stain in the dark at 37°C until bands appear. Rinse in water.

APPENDIX 2 (cont.)

Phosphoglucomutase (PGM)

0.1 M Tris-HCl, pH 8.0	50 ml
_-D-glucose-1-phosphate, disodium salt	50 mg
_-D-glucose-1,6-diphosphate	0.5 mg
1.0 M $MgCl_2$	0.5 ml
NAD	5 mg
Glucose-6-phosphate dehydrogenase	160 units
MTT	5 mg
PMS	1 mg

obs: stain in the dark at room temperature until bands appear. Rinse in water.

Triose-phosphate isomerase (TPI)

0.1 Tris-HCl, pH 8.0	50 ml
Dihydroxyacetone phosphate, lithium salt	5 mg
EDTA, tetrasodium salt, dihydrate	19 mg
NAD	15 mg
MTT	5 mg
PMS	1 mg
Arsenic acid, sodium salt	230 mg
Glyceraldehyd-3-phosphate dehydrogenase	300 units

obs: stain in the dark at 37°C until bands appear. Rinse in water.

6-phosphogluconate dehydrogenase (6PG)

0.1 M Tris-HCl, pH unajusted	50 ml
6-phosphogluconic acid, barium salt	30 mg
EDTA Na_4, dihydrate	20 mg
Combine, and adjust pH to 8.0, then add:	
1.0 M $MgCl_2$	1 ml
NADP	2.5 mg
MTT	4 mg
PMS	1 mg

obs: stain in the dark at 37°C until bands appear. Rinse in water.

APPENDIX 3

Total number of isolates in trees and seedlings, at **high** várzea, during four evaluation periods.

	-------TREES--------					-------SEEDLINGS-------				
TAXON	I	II	III	IV	T	I	II	III	IV	T
A. elaeidicola	1	0	0	0	**1**	0	0	0	0	**0**
Arthroconidial	3	1	0	1	**5**	3	2	0	0	**5**
C. gloesporioides	7	3	3	1	**14**	5	7	0	3	**15**
H. serpens	4	15	6	14	**39**	1	29	4	18	**52**
H. stygium	1	3	7	14	**25**	4	4	7	14	**29**
I. euterpes	3	1	0	0	**4**	1	1	1	1	**4**
Letendraea sp.	3	7	3	9	**22**	12	86	11	17	**126**
Phoma sp.	2	4	1	1	**8**	0	1	1	2	**4**
Phomopsis sp.	15	8	9	7	**39**	10	7	2	6	**25**
Thozetella sp.	2	0	1	2	**5**	1	7	8	9	**25**
U. deusta	0	7	1	0	**8**	0	19	5	2	**26**
X. adscendens	10	5	38	6	**59**	0	8	8	6	**22**
X. arbuscula	2	4	34	15	**55**	1	6	29	23	**59**
X. cubensis	28	82	61	53	**224**	26	63	101	90	**280**
X. curta	0	1	3	33	**37**	0	2	2	71	**75**
X. multiplex	0	2	9	4	**15**	3	3	6	8	**20**
Xylariaceae I	2	5	6	2	**15**	1	7	4	5	**17**
Xylariaceae II	3	0	2	2	**7**	1	5	11	4	**21**
Xylariaceae III	0	6	8	3	**17**	2	12	11	11	**36**
Xylariaceae IV	6	3	0	3	**12**	1	5	6	3	**15**
Xylariaceae V	3	0	10	2	**15**	2	9	3	0	**14**

APPENDIX 4

Total number of isolates in trees and seedlings, at **low** várzea, during four evaluation periods.

	-------TREES-------					-----SEEDLINGS-----				
TAXON	I	II	III	IV	**T**	I	II	III	IV	**T**
A. elaeidicola	4	0	1	0	**5**	1	4	0	15	**20**
Arthroconidial	7	10	6	5	**28**	2	4	1	0	**7**
C. gloesporioides	6	1	5	0	**12**	11	6	0	0	**17**
H. serpens	3	1	0	6	**10**	7	8	2	29	**46**
H. stygium	3	3	1	3	**12**	8	4	11	18	**41**
I. euterpes	16	2	1	8	**27**	12	7	3	4	**26**
Letendraea sp.	5	7	0	9	**21**	101	111	35	64	**311**
Phoma sp.	7	3	3	2	**15**	8	0	1	1	**10**
Phomopsis sp. 1	5	1	5	14	**25**	5	1	0	2	**8**
Thozetella sp.	1	0	0	1	**2**	16	32	13	32	**93**
U. deusta	0	0	0	0	**0**	0	0	0	2	**2**
X. adscendens	7	1	0	0	**8**	10	3	13	1	**27**
X. arbuscula	2	0	3	0	**5**	4	3	28	4	**39**
X. cubensis	42	1	12	8	**63**	35	9	35	25	**104**
X. curta	0	0	0	2	**2**	1	3	3	1	**8**
X. multiplex	10	0	2	0	**12**	1	0	10	2	**13**
Xylariaceae I	2	0	0	2	**4**	6	12	8	34	**60**
Xylariaceae II	12	0	0	1	**13**	8	13	8	1	**30**
Xylariaceae III	9	0	0	3	**12**	6	6	9	8	**29**
Xylariaceae IV	8	0	1	0	**9**	10	6	2	1	**19**
Xylariaceae V	5	1	1	0	**7**	5	2	5	1	**13**

APPENDIX 5

Character state matrix for *Xylaria cubensis.*

1: isozyme phenotype, 2-7: colony characteristics,
8-10: stromata features, 11: conidium size.

	CHARACTERS										
STRAINS	***1***	***2***	***3***	***4***	***5***	***6***	***7***	***8***	***9***	***10***	***11***
3	4	1	4	2	0	1	0	1	1	1	1
8	1	1	4	2	0	1	0	3	3	1	1
23	9	1	4	2	1	1	0	1	3	1	1
29	14	1	4	2	0	1	0	1	1	1	2
101	1	2	1	3	0	1	1	1	3	2	2
102	2	1	1	2	1	2	1	1	1	2	2
103	3	2	3	1	1	1	1	1	1	2	2
104	1	1	3	2	1	1	0	2	2	1	1
105	1	1	4	2	1	1	0	1	1	1	1
106	5	1	1	2	1	2	1	1	1	2	2
107	6	2	3	1	1	2	1	1	3	2	2
108	7	1	4	2	1	1	0	3	3	1	1
109	1	1	2	2	1	2	1	3	3	1	1
110	8	2	1	2	1	2	1	3	3	2	1
112	1	1	4	2	0	1	0	1	1	1	1
113	1	1	1	1	1	2	1	1	1	2	1
114	1	1	2	3	1	1	1	3	3	2	1
115	6	1	4	1	0	1	0	3	3	2	2
116	1	1	4	1	0	1	0	2	2	1	1
117	6	1	2	2	0	1	0	1	3	1	2
118	10	1	2	2	1	2	1	3	3	2	2
119	1	1	3	2	1	1	0	2	2	1	1
120	6	1	2	1	0	1	0	3	3	1	1
121	11	2	2	1	0	1	0	1	1	1	1
122	12	1	4	2	0	1	0	1	1	1	1
123	13	1	4	2	1	2	2	2	2	2	1
124	10	1	4	1	0	1	0	3	3	1	1
125	14	1	4	1	0	1	0	1	1	1	2
126	6	1	2	1	0	1	1	3	3	2	1
127	1	1	4	1	0	1	0	1	1	2	1
128	11	1	1	2	1	2	1	1	1	2	2
129	10	1	4	1	0	1	0	3	3	1	1
130	15	1	2	2	0	1	1	1	1	2	2
131	1	1	4	2	1	1	0	1	1	1	1
132	15	2	2	2	0	1	0	1	1	1	2

APPENDIX 6

Pairing reactions among 35 strains of *X. cubensis* subgroup 1 (a), subgroup 2 (b), and subgroup 3 (c). 0= somatic incompatibility, 1= somatic compatibility.

(a)

	2	5	7	10	12	14	15	19	21	24	29	34
2	1											
5	1	1										
7	1	1	1									
10	0	1	0	1								
12	0	1	0	1	1							
14	1	0	1	1	0	1						
15	0	0	0	0	1	1	1					
19	1	1	1	1	1	1	1	1				
21	1	1	1	0	0	1	0	1	1			
24	0	0	0	0	1	1	0	1	0	1		
29	0	1	1	1	0	1	1	1	1	1	1	
34	0	0	0	0	0	0	1	0	0	0	1	1

(b)

	1	3	4	6	8	13	16	17	22	23	26	27	30	31	33	35
1	1															
3	1	1														
4	0	0	1													
6	0	1	0	1												
8	1	0	0	0	1											
13	0	0	0	0	0	1										
16	0	0	0	0	0	0	1									
17	0	1	0	0	0	1	0	1								
22	1	1	0	1	1	0	1	0	1							
23	0	1	0	1	1	1	1	1	1	1						
26	0	1	0	1	0	1	0	1	0	1	1					
27	0	0	0	0	0	0	0	0	1	1	1	1				
30	0	0	0	0	0	0	1	0	0	1	0	0	1			
31	0	1	0	0	0	0	0	1	0	0	0	0	0	1		
33	0	0	0	0	1	0	0	0	0	1	1	0	0	0	1	
35	0	0	1	1	0	1	0	0	0	1	1	0	0	1	1	1

(c)

	9	11	18	20	25	28	32
9	1						
11	0	1					
18	1	0	1				
20	0	0	0	1			
25	0	1	1	0	1		
28	1	1	0	0	1	1	
32	0	1	1	1	1	1	1

APPENDIX 7

Results of the ANOVA Test for the effect of leaf age, leaf tissue, plant developmental stage, season and subsite on the overall leaf colonization by the endophytic fungi.

Source of var.	**DF**	**MS**	**F**	**P**
Leaf age	2	4925.9	161.89	< 0.001
Leaf tissue	1	1106.8	36.37	< 0.001
Plant stage	1	1669.6	54.87	< 0.001
Season	1	34.4	1.13	n.s.
Subsite	1	649.0	21.33	< 0.001
Error	437	30.4		

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